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NEWS 1	Web Page URLs for STN Seminar Schedule - N. America
NEWS 2	Apr 08 "Ask CAS" for self-help around the clock
NEWS 3	Apr 09 BEILSTEIN: Reload and Implementation of a New Subject Area
NEWS 4	Apr 09 ZDB will be removed from STN
NEWS 5	Apr 19 US Patent Applications available in IFICDB, IFIPAT, and IFIUDB
NEWS 6	Apr 22 Records from IP.com available in CAPLUS, HCAPLUS, and ZCAPLUS
NEWS 7	Apr 22 BIOSIS Gene Names now available in TOXCENTER
NEWS 8	Apr 22 Federal Research in Progress (FEDRIP) now available
NEWS 9	Jun 03 New e-mail delivery for search results now available
NEWS 10	Jun 10 MEDLINE Reload
NEWS 11	Jun 10 PCTFULL has been reloaded
NEWS 12	Jul 02 FOREGE no longer contains STANDARDS file segment
NEWS 13	Jul 22 USAN to be reloaded July 28, 2002; saved answer sets no longer valid
NEWS 14	Jul 29 Enhanced polymer searching in REGISTRY
NEWS 15	Jul 30 NETFIRST to be removed from STN
NEWS 16	Aug 08 CANCERLIT reload
NEWS 17	Aug 08 PHARMAMarketLetter(PHARMAML) - new on STN
NEWS 18	Aug 08 NTIS has been reloaded and enhanced
NEWS 19	Aug 19 Aquatic Toxicity Information Retrieval (AQUIRE) now available on STN
NEWS 20	Aug 19 IFIPAT, IFICDB, and IFIUDB have been reloaded
NEWS 21	Aug 19 The MEDLINE file segment of TOXCENTER has been reloaded
NEWS 22	Aug 26 Sequence searching in REGISTRY enhanced
NEWS 23	Sep 03 JAPIO has been reloaded and enhanced
NEWS 24	Sep 16 Experimental properties added to the REGISTRY file
NEWS 25	Sep 16 CA Section Thesaurus available in CAPLUS and CA
NEWS 26	Oct 01 CASREACT Enriched with Reactions from 1907 to 1985
NEWS 27	Oct 21 EVENTLINE has been reloaded
NEWS 28	Oct 24 BEILSTEIN adds new search fields
NEWS 29	Oct 24 Nutraceuticals International (NUTRACEUT) now available on STN
NEWS 30	Oct 25 MEDLINE SDI run of October 8, 2002
NEWS 31	Nov 18 DKILIT has been renamed APOLLIT
NEWS 32	Nov 25 More calculated properties added to REGISTRY
NEWS 33	Dec 02 TIBKAT will be removed from STN
NEWS 34	Dec 04 CSA files on STN
NEWS 35	Dec 17 PCTFULL now covers WP/PCT Applications from 1978 to date
NEWS 36	Dec 17 TOXCENTER enhanced with additional content
NEWS 37	Dec 17 Adis Clinical Trials Insight now available on STN
NEWS 38	Dec 30 ISMEC no longer available
NEWS 39	Jan 21 NUTRACEUT offering one free connect hour in February 2003
NEWS 40	Jan 21 PHARMAML offering one free connect hour in February 2003
NEWS 41	Jan 29 Simultaneous left and right truncation added to COMPENDEX, ENERGY, INSPEC
NEWS 42	Feb 13 CANCERLIT is no longer being updated
NEWS 43	Feb 24 METADEX enhancements
NEWS 44	Feb 24 PCTGEN now available on STN
NEWS 45	Feb 24 TEMA now available on STN

NEWS 46 Feb 26 NTIS now allows simultaneous left and right truncation
NEWS 47 Feb 26 PCTFULL now contains images
NEWS 48 Mar 04 SDI PACKAGE for monthly delivery of multifile SDI results
NEWS 49 Mar 19 APOLLIT offering free connect time in April 2003
NEWS 50 Mar 20 EVENTLINE will be removed from STN
NEWS 51 Mar 24 PATDPAFULL now available on STN
NEWS 52 Mar 24 Additional information for trade-named substances without structures available in REGISTRY
NEWS 53 Mar 24 Indexing from 1957 to 1966 added to records in CA/CAPLUS

NEWS EXPRESS January 6 CURRENT WINDOWS VERSION IS V6.01a,
CURRENT MACINTOSH VERSION IS V6.0b(ENG) AND V6.0Jb(JP),
AND CURRENT DISCOVER FILE IS DATED 01 OCTOBER 2002

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=> file medline, uspatful, dgene, embase, wpids, fsta, jicst
COST IN U.S. DOLLARS SINCE FILE TOTAL
ENTRY SESSION
FULL ESTIMATED COST 0.21 0.21

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FILE 'USPATFULL' ENTERED AT 15:42:44 ON 28 MAR 2003
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BILIPROTEIN IS NOT A RECOGNIZED COMMAND
The previous command name entered was not recognized by the system.
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7544

J

"HELP COMMANDS" at an arrow prompt (=>).

=> s biliprotein
L1 177 BILIPROTEIN

=> s l1 and fusion protein
L2 3 L1 AND FUSION PROTEIN

=> d l2 ti abs ibib tot

L2 ANSWER 1 OF 3 USPATFULL

TI PHYTOFLUORS AS FLUORESCENT LABELS

AB This invention provides new fluorescent molecules useful for detection of target entities. In particular, it relates to fluorescent adducts comprising an apoprotein and a bilin.

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

ACCESSION NUMBER: 2002:37522 USPATFULL

TITLE: PHYTOFLUORS AS FLUORESCENT LABELS

INVENTOR(S): LAGARIAS, JOHN CLARK, DAVIS, CA, UNITED STATES

	NUMBER	KIND	DATE
PATENT INFORMATION:	US 2002022239	A1	20020221
APPLICATION INFO.:	US 1999-272809	A1	19990319 (9)
DOCUMENT TYPE:	Utility		
FILE SEGMENT:	APPLICATION		
LEGAL REPRESENTATIVE:	LAW OFFICES OF JONATHAN ALAN QUINE, PO BOX 458, ALAMEDA, CA, 94501		
NUMBER OF CLAIMS:	32		
EXEMPLARY CLAIM:	1		
NUMBER OF DRAWINGS:	3 Drawing Page(s)		
LINE COUNT:	2727		

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

L2 ANSWER 2 OF 3 USPATFULL

TI Recombinant phycobiliprotein and phycobiliprotein linker fusion proteins and uses therefore

AB This invention is directed to the utilization of the developing methods for molecular manipulation of cyanobacteria and red algae (and potentially cryptomonad algae) to express of phycobiliproteins and phycobiliprotein linker fusion proteins and their utilization as phycobiliprotein, phycobilisome and subassembly based reagents. In particular, the present invention relates to a method for a specific binding assay to determine a target moiety which is a member of a specific binding pair, and provides an improvement in the method comprising using a detectable label which is a **fusion protein** containing both a phycobiliprotein domain and another domain corresponding to a first member of a specific binding pair, where the **fusion protein** binds to a second member of the specific binding pair to provide a detectable labeled complex. The domain derived from the first member of the specific binding pair can be directly fused to the phycobiliprotein or phycobiliprotein linker domain or be separated by a spacer that allows correct folding of both domains.

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

ACCESSION NUMBER: 2001:237667 USPATFULL

TITLE: Recombinant phycobiliprotein and phycobiliprotein linker fusion proteins and uses therefore

INVENTOR(S): Allnutt, F.C. Thomas, Port Deposit, MD, United States
Toole, Colleen Mary, New Winson, MD, United States
Morseman, John Peter, Columbia, MD, United States

NUMBER	KIND	DATE
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PATENT INFORMATION: US 2001055783 A1 20011227
APPLICATION INFO.: US 2001-882093 A1 20010618 (9)

NUMBER DATE

PRIORITY INFORMATION: US 2000-211784P 20000616 (60)
DOCUMENT TYPE: Utility
FILE SEGMENT: APPLICATION
LEGAL REPRESENTATIVE: BROBECK, PHLEGER & HARRISON, LLP, ATTN: INTELLECTUAL PROPERTY DEPARTMENT, 1333 H STREET, N.W. SUITE 800, WASHINGTON, DC, 20005
NUMBER OF CLAIMS: 46
EXEMPLARY CLAIM: 1
LINE COUNT: 1218
CAS INDEXING IS AVAILABLE FOR THIS PATENT.

L2 ANSWER 3 OF 3 USPATFULL
TI Phytofluors as fluorescent labels
AB This invention provides new fluorescent molecules useful for detection of target entities. In particular, it relates to fluorescent adducts comprising an apoprotein and a bilin.

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

ACCESSION NUMBER: 2000:40859 USPATFULL
TITLE: Phytofluors as fluorescent labels
INVENTOR(S): Lagarias, John Clark, Davis, CA, United States
Murphy, John Thomas, San Francisco, CA, United States
PATENT ASSIGNEE(S): The Regents of the University of California, Oakland, CA, United States (U.S. corporation)

NUMBER KIND DATE

PATENT INFORMATION: US 6046014 20000404
APPLICATION INFO.: US 1997-904871 19970801 (8)

NUMBER DATE

PRIORITY INFORMATION: US 1996-23217P 19960802 (60)
DOCUMENT TYPE: Utility
FILE SEGMENT: Granted
PRIMARY EXAMINER: Houtteman, Scott W.
LEGAL REPRESENTATIVE: Majestic, Parsons, Siebert & Hsue, P.C.
NUMBER OF CLAIMS: 27
EXEMPLARY CLAIM: 1
NUMBER OF DRAWINGS: 37 Drawing Figure(s); 14 Drawing Page(s)
LINE COUNT: 3666
CAS INDEXING IS AVAILABLE FOR THIS PATENT.

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(FILE 'HOME' ENTERED AT 15:42:24 ON 28 MAR 2003)

FILE 'MEDLINE, USPATFULL, DGENE, EMBASE, WPIDS, FSTA, JICST-EPLUS'
ENTERED AT 15:42:44 ON 28 MAR 2003

L1 177 S BILIPROTEIN
L2 3 S L1 AND FUSION PROTEIN

=> s l1 and holo alpha subunit
L3 0 L1 AND HOLO ALPHA SUBUNIT

=> s l1 and apo
L4 1 L1 AND APO

=> d 14 ti abs ibib tot

L4 ANSWER 1 OF 1 USPATFULL
TI Phytofluors as fluorescent labels
AB This invention provides new fluorescent molecules useful for detection of target entities. In particular, it relates to fluorescent adducts comprising an apoprotein and a bilin.

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

ACCESSION NUMBER: 2000:40859 USPATFULL
TITLE: Phytofluors as fluorescent labels
INVENTOR(S): Lagarias, John Clark, Davis, CA, United States
PATENT ASSIGNEE(S): Murphy, John Thomas, San Francisco, CA, United States
The Regents of the University of California, Oakland, CA, United States (U.S. corporation)

	NUMBER	KIND	DATE
PATENT INFORMATION:	US 6046014		20000404
APPLICATION INFO.:	US 1997-904871		19970801 (8)

	NUMBER	DATE
PRIORITY INFORMATION:	US 1996-23217P	19960802 (60)
DOCUMENT TYPE:	Utility	
FILE SEGMENT:	Granted	
PRIMARY EXAMINER:	Houtteman, Scott W.	
LEGAL REPRESENTATIVE:	Majestic, Parsons, Siebert & Hsue, P.C.	
NUMBER OF CLAIMS:	27	
EXEMPLARY CLAIM:	1	
NUMBER OF DRAWINGS:	37 Drawing Figure(s); 14 Drawing Page(s)	
LINE COUNT:	3666	

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

=> d his

(FILE 'HOME' ENTERED AT 15:42:24 ON 28 MAR 2003)

FILE 'MEDLINE, USPATFULL, DGENE, EMBASE, WPIDS, FSTA, JICST-EPLUS'
ENTERED AT 15:42:44 ON 28 MAR 2003

L1 177 S BILIPROTEIN
L2 3 S L1 AND FUSION PROTEIN
L3 0 S L1 AND HOLO ALPHA SUBUNIT
L4 1 S L1 AND APO

=> s l1 and holo
L5 0 L1 AND HOLO

=> s l1 and heme
L6 5 L1 AND HEME

=> d 16 ti abs ibib tot

L6 ANSWER 1 OF 5 MEDLINE
TI Developmental profile, isolation, and biochemical characterization of a novel lipoglycoheme-carrier protein from the American dog tick, *Dermacentor variabilis* (Acari: Ixodidae) and observations on a similar protein in the soft tick, *Ornithodoros parkeri* (Acari: Argasidae).
AB A novel lipoglycoheme-carrier protein (CP) in the American dog tick, *Dermacentor variabilis* (Say) has been purified and characterized. CP was purified by native-PAGE from partially fed virgin females. CP has a density of 1.25 g/ml with a molecular weight of 200 K by native-PAGE and

340 K by gel filtration chromatography. CP is comprised of two major subunits, 98 K and 92 K in molecular weight by SDS-PAGE. Separate amino acid composition of the two subunits indicated high contents of As(x), Gl(x) and leucine. However, the N-terminal amino acid sequence of the two subunits was only 13% identical. The lower molecular weight subunit showed 61% identity to artemocyanin (**biliprotein**) in fairy shrimps, 46% identity to minor vitellogenin in chickens and 13% identity to vitellin of the black-legged tick. No similarity match was found for the other subunit. CP is a lipoglycoheme-protein as indicated by selective staining of native-PAGE gel for lipids, carbohydrates and **heme**. Lipid analysis by thin layer chromatography revealed the presence of cholesterol, phospholipids, monoacylglycerides, triacylglycerides and free fatty acids. **Heme** associated with purified CP demonstrated a lambda(max) of 397.5 nm while the lambda(max) of crude hemolymph plasma was 402.5 nm. The presence of CP in whole body homogenates of eggs, unfed and fed larvae and fed nymphs as well as in the plasma of unfed and fed adults including vitellogenic females was demonstrated by native-PAGE. Although a protein of analogous size was not found in the soft tick, *Ornithodoros parkeri* Cooley, a high molecular weight protein (500 K) is the predominant plasma protein in both unfed and fed male and female adults of that species as determined by native-PAGE. Also, CP appears to function as a **biliprotein** which sequesters **heme**.

ACCESSION NUMBER: 2001236262 MEDLINE
DOCUMENT NUMBER: 21124779 PubMed ID: 11222939
TITLE: Developmental profile, isolation, and biochemical characterization of a novel lipoglycoheme-carrier protein from the American dog tick, *Dermacentor variabilis* (Acari: Ixodidae) and observations on a similar protein in the soft tick, *Ornithodoros parkeri* (Acari: Argasidae).
AUTHOR: Gudderra N P; Neese P A; Sonenshine D E; Apperson C S; Roe R M
CORPORATE SOURCE: Department of Entomology, North Carolina State University, Raleigh, NC 27695-7647, USA.
CONTRACT NUMBER: 1 RO1 AI 36257 (NIAID)
SOURCE: INSECT BIOCHEMISTRY AND MOLECULAR BIOLOGY, (2001 Mar 15) 31 (4-5) 299-311.
Journal code: 9207282. ISSN: 0965-1748.
PUB. COUNTRY: England: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200105
ENTRY DATE: Entered STN: 20010517
Last Updated on STN: 20010517
Entered Medline: 20010503

L6 ANSWER 2 OF 5 MEDLINE
TI Coupled oxidation of **heme** covalently attached to cytochrome b562 yields a novel **biliprotein**.
AB A variant of *Escherichia coli* cytochrome b(562) with covalently attached **heme** can be converted to a biliverdin-containing protein in two distinct stages by coupled oxidation and acid hydrolysis. The first stage of coupled oxidation yields a stable verdoheme-containing protein. This verdoheme protein is unusual in three respects. First, the verdoheme group is covalently bound to the protein through a c-type thioether linkage. Second, the oxidation stops at the verdoheme stage, and finally, this is the first report of verdoheme generated from a **heme** protein with exclusive methionine ligation to the **heme** iron. In addition, the oxidation process does not require denaturation of the protein. The product has been characterized by optical spectroscopy, ESI mass spectrometry, and (1)H NMR. The NMR data show that the predominant product is the result of oxidation at the alpha-meso carbon. A collective evaluation of data on the topic suggests that the electronic structure of the **heme**, not protein steric effects, is the main factor in

controlling the regiospecificity of the oxidation site. In the second stage of conversion to a **biliprotein**, we demonstrate that the verdoheme ring can be opened by treatment with aqueous formic acid to give alpha-biliverdin covalently attached to the folded protein. This product, a protein-bound linear tetrapyrrole as characterized by optical spectroscopy and mass spectrometry, is an example of a phycobilin chromophore that has not been observed previously.

ACCESSION NUMBER: 2000074535 MEDLINE
DOCUMENT NUMBER: 20074535 PubMed ID: 10606518
TITLE: Coupled oxidation of **heme** covalently attached to cytochrome b562 yields a novel **biliprotein**.
AUTHOR: Rice J K; Fearnley I M; Barker P D
CORPORATE SOURCE: Naval Research Laboratory, Washington, D.C. 20375-5342, USA.
SOURCE: BIOCHEMISTRY, (1999 Dec 21) 38 (51) 16847-56.
Journal code: 0370623. ISSN: 0006-2960.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200001
ENTRY DATE: Entered STN: 20000131
Last Updated on STN: 20000131
Entered Medline: 20000119

L6 ANSWER 3 OF 5 MEDLINE
TI The molecular structure of insecticyanin from the tobacco hornworm *Manduca sexta* L. at 2.6 Å resolution.
AB Insecticyanin, a blue **biliprotein** isolated from the tobacco hornworm *Manduca sexta* L., is involved in insect camouflage. Its three-dimensional structure has now been solved to 2.6 Å resolution using the techniques of multiple isomorphous replacement, non-crystallographic symmetry averaging about a local 2-fold rotation axis and solvent flattening. All 189 amino acids have been fitted to the electron density map. The map clearly shows that insecticyanin is a tetramer with one of its molecular 2-fold axes coincident to a crystallographic dyad. The individual subunits have overall dimensions of 44 Å X 37 Å X 40 Å and consist primarily of an eight-stranded anti-parallel beta-barrel flanked on one side by a 4.5-turn alpha-helix. Interestingly the overall three-dimensional fold of the insecticyanin subunit shows remarkable similarity to the structural motifs of bovine beta-lactoglobulin and the human serum retinol-binding protein. The electron density attributable to the chromophore is unambiguous and shows that it is indeed the gamma-isomer of biliverdin. The biliverdin lies towards the open end of the beta-barrel with its two propionate side chains pointing towards the solvent and it adopts a rather folded conformation, much like a **heme**.

ACCESSION NUMBER: 87275848 MEDLINE
DOCUMENT NUMBER: 87275848 PubMed ID: 3608987
TITLE: The molecular structure of insecticyanin from the tobacco hornworm *Manduca sexta* L. at 2.6 Å resolution.
AUTHOR: Holden H M; Rypniewski W R; Law J H; Rayment I
CONTRACT NUMBER: AM GM 351865 (NIADDK)
BRSG 829023 (DRS)
GM 29238 (NIGMS)
SOURCE: EMBO JOURNAL, (1987 Jun) 6 (6) 1565-70.
Journal code: 8208664. ISSN: 0261-4189.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198709
ENTRY DATE: Entered STN: 19900305
Last Updated on STN: 19970203

Entered Medline: 19870924

L6 ANSWER 4 OF 5 USPATFULL

TI Reduction of oxyradical damage in biomedical applications
AB The biliproteins delta-bilirubin and delta-bilipeptide are useful as a cytoprotective antioxidants. Delta-bilipeptide as the term is used herein is a truncated form of delta-bilirubin in which an albumin analogue of 10-200 amino acid residues replaces the albumin portion of delta-bilirubin. Patient-administrable compositions for addition to a patient's blood to minimize oxyradical damage caused by ischemia-reperfusion injury that may result in various surgical procedures, and comprising delta-bilirubin or delta-bilipeptide, are described.

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

ACCESSION NUMBER: 91:73347 USPATFULL
TITLE: Reduction of oxyradical damage in biomedical applications
INVENTOR(S): Wu, Tai-Wing, Toronto, Canada
PATENT ASSIGNEE(S): Nagase Co., Ltd., Osaka, Japan (non-U.S. corporation)

NUMBER KIND DATE

NUMBER	KIND	DATE
PATENT INFORMATION:	US 5047395	19910910
APPLICATION INFO.:	US 1990-554197	19900717 (7)
DOCUMENT TYPE:	Utility	
FILE SEGMENT:	Granted	
PRIMARY EXAMINER:	Waddell, Frederick E.	
ASSISTANT EXAMINER:	Wilson, Terry	
LEGAL REPRESENTATIVE:	Wyatt, Gerber, Burke and Badie	
NUMBER OF CLAIMS:	20	
EXEMPLARY CLAIM:	1	
NUMBER OF DRAWINGS:	1 Drawing Figure(s); 1 Drawing Page(s)	
LINE COUNT:	430	

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

L6 ANSWER 5 OF 5 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.

TI Coupled oxidation of heme covalently attached to cytochrome b562 yields a novel biliprotein.

AB A variant of Escherichia coli cytochrome b562 with covalently attached heme can be converted to a biliverdin-containing protein in two distinct stages by coupled oxidation and acid hydrolysis. The first stage of coupled oxidation yields a stable verdoheme-containing protein. This verdoheme protein is unusual in three respects. First, the verdoheme group is covalently bound to the protein through a c-type thioether linkage. Second, the oxidation stops at the verdoheme stage, and finally, this is the first report of verdoheme generated from a heme protein with exclusive methionine ligation to the heme iron. In addition, the oxidation process does not require denaturation of the protein. The product has been characterized by optical spectroscopy, ESI mass spectrometry, and ¹H NMR. The NMR data show that the predominant product is the result of oxidation at the .alpha.-meso carbon. A collective evaluation of data on the topic suggests that the electronic structure of the heme, not protein steric effects, is the main factor in controlling the regiospecificity of the oxidation site. In the second stage of conversion to a biliprotein, we demonstrate that the verdoheme ring can be opened by treatment with aqueous formic acid to give .alpha.-biliverdin covalently attached to the folded protein. This product, a protein-bound linear tetrapyrrole as characterized by optical spectroscopy and mass spectrometry, is an example of a phycobilin chromophore that has not been observed previously.

ACCESSION NUMBER: 2000009879 EMBASE

TITLE: Coupled oxidation of heme covalently attached to cytochrome b562 yields a novel biliprotein.

AUTHOR: Rice J.K.; Fearnley I.M.; Barker P.D.
CORPORATE SOURCE: P.D. Barker, Centre for Protein Engineering, MRC Centre,
Hills Road, Cambridge CB2 2QH, United Kingdom.
pxb@mrc-lmb.cam.ac.uk
SOURCE: Biochemistry, (21 Dec 1999) 38/51 (16847-16856).
Refs: 36
ISSN: 0006-2960 CODEN: BICHAW
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English

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saved answer sets no longer valid
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NEWS EXPRESS January 6 CURRENT WINDOWS VERSION IS V6.01a,
CURRENT MACINTOSH VERSION IS V6.0b(ENG) AND V6.0Jb(JP),
AND CURRENT DISCOVER FILE IS DATED 01 OCTOBER 2002

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NEWS WWW CAS World Wide Web Site (general information)

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FILE 'BIOSIS' ENTERED AT 15:17:25 ON 28 MAR 2003
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FILE 'JAPIO' ENTERED AT 15:17:25 ON 28 MAR 2003
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=> s phycoerythrocyanin

=> s phycoerythrocyanin

L1 143 PHYCOERYTHROCYANIN

=> s holo alpha subunit
L2 9 HOLO ALPHA SUBUNIT

=> d 12 and 11
L1 IS NOT VALID HERE
For an explanation, enter "HELP DISPLAY".

=> s 12 and 11
L3 3 L2 AND L1

=> d 12 ti abs ibib tot

L2 ANSWER 1 OF 9 MEDLINE

TI Biosynthesis of the cyanobacterial light-harvesting polypeptide phycoerythrocyanin **holo-alpha subunit** in a heterologous host.

AB The entire pathway for the biosynthesis of the phycobiliviolin-bearing His-tagged **holo-alpha subunit** of the cyanobacterial photosynthetic accessory protein phycoerythrocyanin was reconstituted in Escherichia coli. Cyanobacterial genes encoding enzymes required for the conversion of heme to 3Z-phycocyanobilin, a precursor of phycobiliviolin (namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase), were expressed from a plasmid under the control of the hybrid trp-lac (trc) promoter. Genes for the apo-phycoerythrocyanin alpha subunit (pecA) and the heterodimeric lyase/isomerase (pecE and pecF), which catalyzes both the covalent attachment of phycocyanobilin and its concurrent isomerization to phycobiliviolin, were expressed from the trc promoter on a second plasmid. Upon induction, recombinant E. coli used endogenous heme to produce holo-PecA with absorbance and fluorescence properties similar to those of the same protein produced in cyanobacteria. About two-thirds of the apo-PecA was converted to holo-PecA. No significant bilin addition took place in a similarly engineered E. coli strain that lacks pecE and pecF. By using immobilized metal affinity chromatography, both apo-PecA and holo-PecA were isolated as ternary complexes with PecE and PecF. The identities of all three components in the ternary complexes were established unambiguously by protein and tryptic peptide analyses performed by matrix-assisted laser desorption ionization-time of flight mass spectrometry.

ACCESSION NUMBER: 2002415572 MEDLINE

DOCUMENT NUMBER: 22159919 PubMed ID: 12169589

TITLE: Biosynthesis of the cyanobacterial light-harvesting polypeptide phycoerythrocyanin **holo-alpha subunit** in a heterologous host.

AUTHOR: Tooley Aaron J; Glazer Alexander N

CORPORATE SOURCE: Department of Molecular and Cell Biology, University of California, Berkeley, CA 94720-3200, USA.

SOURCE: JOURNAL OF BACTERIOLOGY, (2002 Sep) 184 (17) 4666-71.
Journal code: 2985120R. ISSN: 0021-9193.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200208

ENTRY DATE: Entered STN: 20020810

Last Updated on STN: 20020830

Entered Medline: 20020829

Valid date

L2 ANSWER 2 OF 9 MEDLINE

TI Biosynthesis of a fluorescent cyanobacterial C-phycocyanin **holo-alpha subunit** in a heterologous host.

AB The entire pathway for the synthesis of a fluorescent holophycobiliprotein

subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803) was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to the natural chromophore 3Z-phycocyanobilin, namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase, were expressed from a plasmid under control of the hybrid trp-lac (trc) promoter. Genes for the apoprotein (C-phycocyanin alpha subunit; cpcA) and the heterodimeric lyase (cpcE and cpcF) that catalyzes chromophore attachment were expressed from the trc promoter on a second plasmid. Upon induction, recombinant *E. coli* used the cellular pool of heme to produce holo-CpcA with spectroscopic properties qualitatively and quantitatively similar to those of the same protein produced endogenously in cyanobacteria. About a third of the apo-CpcA was converted to holo-CpcA. No significant bilin addition took place in a similarly engineered *E. coli* strain that lacks cpcE and cpcF. This approach should permit incisive analysis of many remaining questions in phycobiliprotein biosynthesis. These studies also demonstrate the feasibility of generating constructs of these proteins *in situ* for use as fluorescent protein probes in living cells.

ACCESSION NUMBER: 2001504133 MEDLINE
DOCUMENT NUMBER: 21438034 PubMed ID: 11553806
TITLE: Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-alpha subunit in a heterologous host.
AUTHOR: Tooley A J; Cai Y A; Glazer A N
CORPORATE SOURCE: Department of Molecular and Cell Biology, University of California, 142 LSA no. 3200, Berkeley, CA 94720-3200, USA.
SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (2001 Sep 11) 98 (19) 10560-5.
Journal code: 7505876. ISSN: 0027-8424.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200111
ENTRY DATE: Entered STN: 20010913
Last Updated on STN: 20011105
Entered Medline: 20011101

L2 ANSWER 3 OF 9 MEDLINE
TI Phycocyanin alpha-subunit phycocyanobilin lyase.
AB Phycobiliproteins, unlike other light-harvesting proteins involved in photosynthesis, bear covalently attached chromophores. The bilin chromophores are attached through thioether bonds to cysteine residues. The cyanobacterium *Synechococcus* sp. PCC 7002 has eight distinct bilin attachment sites on seven polypeptides, all of which carry the same chromophore, phycocyanobilin. When two genes in the phycocyanin operon of this organism, cpcE and cpcF, are inactivated by insertion, together or separately, the surprising result is elimination of correct bilin attachment at only one site, that on the alpha subunit of phycocyanin. We have overproduced CpcE and CpcF in *Escherichia coli*. In vitro, these proteins catalyze the attachment of phycocyanobilin to the alpha subunit of apophycocyanin at the appropriate site, alpha-Cys-84, to form the correct adduct. CpcE and CpcF also efficiently catalyze the reverse reaction, in which the bilin from holo-alpha subunit is transferred either to the apo-alpha subunit of the same C-phycocyanin or to the apo-alpha subunit of a heterologous C-phycocyanin. The forward and reverse reactions each require both CpcE and CpcF and are specific for the alpha-Cys-84 position. Phycocyanobilin is the immediate precursor of the protein-bound bilin.

ACCESSION NUMBER: 92357762 MEDLINE
DOCUMENT NUMBER: 92357762 PubMed ID: 1495995
TITLE: Phycocyanin alpha-subunit phycocyanobilin lyase.
AUTHOR: Fairchild C D; Zhao J; Zhou J; Colson S E; Bryant D A;
Glazer A N

CORPORATE SOURCE: Department of Molecular and Cell Biology, University of California, Berkeley 94720.
CONTRACT NUMBER: GM28994 (NIGMS)
GM31625 (NIGMS)
SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (1992 Aug 1) 89 (15) 7017-21.
Journal code: 7505876. ISSN: 0027-8424.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199209
ENTRY DATE: Entered STN: 19920925
Last Updated on STN: 19970203
Entered Medline: 19920904

L2 ANSWER 4 OF 9 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
TI Biosynthesis of the cyanobacterial light-harvesting polypeptide phycoerythrocyanin **holo-alpha subunit** in a heterologous host.
AB The entire pathway for the biosynthesis of the phycobiliviolin-bearing His-tagged **holo-alpha subunit** of the cyanobacterial photosynthetic accessory protein phycoerythrocyanin was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to 3Z-phycocyanobilin, a precursor of phycobiliviolin (namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase), were expressed from a plasmid under the control of the hybrid trp-lac (trc) promoter. Genes for the apo-phycoerythrocyanin alpha subunit (pecA) and the heterodimeric lyase/isomerase (pecE and pecF), which catalyzes both the covalent attachment of phycocyanobilin and its concurrent isomerization to phycobiliviolin, were expressed from the trc promoter on a second plasmid. Upon induction, recombinant *E. coli* used endogenous heme to produce holo-PecA with absorbance and fluorescence properties similar to those of the same protein produced in cyanobacteria. About two-thirds of the apo-PecA was converted to holo-PecA. No significant bilin addition took place in a similarly engineered *E. coli* strain that lacks pecE and pecF. By using immobilized metal affinity chromatography, both apo-PecA and holo-PecA were isolated as ternary complexes with PecE and PecF. The identities of all three components in the ternary complexes were established unambiguously by protein and tryptic peptide analyses performed by matrix-assisted laser desorption ionization-time of flight mass spectrometry.

ACCESSION NUMBER: 2002:464629 BIOSIS
DOCUMENT NUMBER: PREV200200464629
TITLE: Biosynthesis of the cyanobacterial light-harvesting polypeptide phycoerythrocyanin **holo-alpha subunit** in a heterologous host.
AUTHOR(S): Tooley, Aaron J.; Glazer, Alexander N. (1)
CORPORATE SOURCE: (1) Natural Reserve System, University of California, 1111 Franklin Street, 6th Floor, Oakland, CA, 94607-5200:
glazer@uclink4.berkeley.edu USA
SOURCE: Journal of Bacteriology, (September, 2002) Vol. 184, No. 17, pp. 4666-4671. <http://intl-jb.asm.org/>. print.
ISSN: 0021-9193.
DOCUMENT TYPE: Article
LANGUAGE: English

L2 ANSWER 5 OF 9 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
TI Biosynthesis of a fluorescent cyanobacterial C-phycoeryanin **holo-alpha subunit** in a heterologous host.
AB The entire pathway for the synthesis of a fluorescent holophycobiliprotein subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803) was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding

enzymes required for the conversion of heme to the natural chromophore 3Z-phycocyanobilin, namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase, were expressed from a plasmid under control of the hybrid trp-lac (trc) promoter. Genes for the apoprotein (C-phycocyanin alpha subunit; cpcA) and the heterodimeric lyase (cpcE and cpcF) that catalyzes chromophore attachment were expressed from the trc promoter on a second plasmid. Upon induction, recombinant E. coli used the cellular pool of heme to produce holo-CpcA with spectroscopic properties qualitatively and quantitatively similar to those of the same protein produced endogenously in cyanobacteria. About a third of the apo-CpcA was converted to holo-CpcA. No significant bilin addition took place in a similarly engineered E. coli strain that lacks cpcE and cpcF. This approach should permit incisive analysis of many remaining questions in phycobiliprotein biosynthesis. These studies also demonstrate the feasibility of generating constructs of these proteins *in situ* for use as fluorescent protein probes in living cells.

ACCESSION NUMBER: 2001:482056 BIOSIS
DOCUMENT NUMBER: PREV200100482056
TITLE: Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-alpha subunit in a heterologous host.
AUTHOR(S): Tooley, Aaron J.; Cai, Yuping A.; Glazer, Alexander N. (1)
CORPORATE SOURCE: (1) Natural Reserve System, University of California System, 1111 Franklin Street, 6th Floor, Oakland, CA, 94607-5200: alexander.glazer@ucop.edu USA
SOURCE: Proceedings of the National Academy of Sciences of the United States of America, (September 11, 2001) Vol. 98, No. 19, pp. 10560-10565. print.
ISSN: 0027-8424.
DOCUMENT TYPE: Article
LANGUAGE: English
SUMMARY LANGUAGE: English

L2 ANSWER 6 OF 9 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
TI PHYCOCYANIN ALPHA-SUBUNIT PHYCOCYANOBILIN LYASE.
AB Phycobiliproteins, unlike other light-harvesting proteins involved in photosynthesis, bear covalently attached chromophores. The bilin chromophores are attached through thioether bonds to cysteine residues. The cyanobacterium *Synechococcus* sp. PCC 7002 has eight distinct bilin attachment sites on seven polypeptides, all of which carry the same chromophore, phycocyanobilin. When two genes in the phycocyanin operon of this organism, cpcE and cpcF, are inactivated by insertion, together or separately, the surprising result is elimination of correct bilin attachment at only one site, that on the .alpha. subunit of phycocyanin. We have overproduced CpcE and CpcF in *Escherichia coli*. In vitro, these proteins catalyze the attachment of phycocyanobilin to the .alpha. subunit of apophycocyanin at the appropriate site, .alpha. Cys-84, to form the correct adduct. CpcE and CpcF also efficiently catalyze the reverse reaction, in which the bilin from holo-.alpha. subunit is transferred either to the apo-.alpha. subunit of the same C-phycocyanin or to the apo-.alpha. subunit of a heterologous C-phycocyanin. The forward and reverse reactions each require both CpcE and CpcF and are specific for the .alpha.-Cys-84 position. Phycocyanobilin is the immediate precursor of the protein-bound bilin.

ACCESSION NUMBER: 1992:506225 BIOSIS
DOCUMENT NUMBER: BA94:124750
TITLE: PHYCOCYANIN ALPHA-SUBUNIT PHYCOCYANOBILIN LYASE.
AUTHOR(S): FAIRCHILD C D; ZHAO J; ZHOU J; COLSON S E; BRYANT D A; GLAZER A N
CORPORATE SOURCE: MCB: STANLEY/DONNER ASU, 229 STANLEY HALL, UNIV. CALIF., BERKELEY, CALIF. 94720.
SOURCE: PROC NATL ACAD SCI U S A, (1992) 89 (15), 7017-7021.
CODEN: PNASA6. ISSN: 0027-8424.
FILE SEGMENT: BA; OLD

LANGUAGE: English

L2 ANSWER 7 OF 9 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.
TI Biosynthesis of the cyanobacterial light-harvesting polypeptide phycoerythrocyanin **holo-.alpha. subunit** in a heterologous host.
AB The entire pathway for the biosynthesis of the phycobiliviolin-bearing His-tagged **holo-.alpha. subunit** of the cyanobacterial photosynthetic accessory protein phycoerythrocyanin was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to 3Z-phycocyanobilin, a precursor of phycobiliviolin (namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase), were expressed from a plasmid under the control of the hybrid trp-lac (trc) promoter. Genes for the apo-phycoerythrocyanin .alpha. subunit (pecA) and the heterodimeric lyase/isomerase (pecE and pecF), which catalyzes both the covalent attachment of phycocyanobilin and its concurrent isomerization to phycobiliviolin, were expressed from the trc promoter on a second plasmid. Upon induction, recombinant *E. coli* used endogenous heme to produce holo-PecA with absorbance and fluorescence properties similar to those of the same protein produced in cyanobacteria. About two-thirds of the apo-PecA was converted to holo-PecA. No significant bilin addition took place in a similarly engineered *E. coli* strain that lacks pecE and pecF. By using immobilized metal affinity chromatography, both apo-PecA and holo-PecA were isolated as ternary complexes with PecE and PecF. The identities of all three components in the ternary complexes were established unambiguously by protein and tryptic peptide analyses performed by matrix-assisted laser desorption ionization-time of flight mass spectrometry.

ACCESSION NUMBER: 2002295599 EMBASE

TITLE: Biosynthesis of the cyanobacterial light-harvesting polypeptide phycoerythrocyanin **holo-.alpha. subunit** in a heterologous host.

AUTHOR: Tooley A.J.; Glazer A.N.

CORPORATE SOURCE: A.N. Glazer, Natural Reserve System, University of California, 1111 Franklin Street, Oakland, CA 94607-5200, United States. glazer@uclink4.berkeley.edu

SOURCE: Journal of Bacteriology, (2002) 184/17 (4666-4671).

Refs: 22

ISSN: 0021-9193 CODEN: JOBAAY

COUNTRY: United States

DOCUMENT TYPE: Journal; Article

FILE SEGMENT: 004 Microbiology

LANGUAGE: English

SUMMARY LANGUAGE: English

L2 ANSWER 8 OF 9 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.

TI Biosynthesis of a fluorescent cyanobacterial C-phycocyanin **holo-.alpha. subunit** in a heterologous host.

AB The entire pathway for the synthesis of a fluorescent holophycobiliprotein subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803) was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to the natural chromophore 3Z-phycocyanobilin, namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase, were expressed from a plasmid under control of the hybrid trp-lac (trc) promoter. Genes for the apoprotein (C-phycocyanin .alpha. subunit; cpcA) and the heterodimeric lyase (cpcE and cpcF) that catalyzes chromophore attachment were expressed from the trc promoter on a second plasmid. Upon induction, recombinant *E. coli* used the cellular pool of heme to produce holo-CpcA with spectroscopic properties qualitatively and quantitatively similar to those of the same protein produced endogenously in cyanobacteria. About a third of the apo-CpcA was converted to holo-CpcA. No significant bilin addition took place in a similarly engineered *E. coli* strain that lacks cpcE and

cpcF. This approach should permit incisive analysis of many remaining questions in phycobiliprotein biosynthesis. These studies also demonstrate the feasibility of generating constructs of these proteins *in situ* for use as fluorescent protein probes in living cells.

ACCESSION NUMBER: 2001329835 EMBASE
TITLE: Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-.alpha. subunit in a heterologous host.
AUTHOR: Tooley A.J.; Cai Y.A.; Glazer A.N.
CORPORATE SOURCE: A.N. Glazer, Natural Reserve System, University of California System, 1111 Franklin Street, Oakland, CA 94607-5200, United States. alexander.glazer@ucop.edu
SOURCE: Proceedings of the National Academy of Sciences of the United States of America, (11 Sep 2001) 98/19 (10560-10565).
Refs: 30
ISSN: 0027-8424 CODEN: PNASA6
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English

L2 ANSWER 9 OF 9 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.
TI Phycocyanin .alpha.-subunit phycocyanobilin lyase.
AB Phycobiliproteins, unlike other light-harvesting proteins involved in photosynthesis, bear covalently attached chromophores. The bilin chromophores are attached through thioether bonds to cysteine residues. The cyanobacterium *Synechococcus* sp. PCC 7002 has eight distinct bilin attachment sites on seven polypeptides, all of which carry the same chromophore, phycocyanobilin. When two genes in the phycocyanin operon of this organism, cpcE and cpcF, are inactivated by insertion, together or separately, the surprising result is elimination of correct bilin attachment at only one site, that on the .alpha. subunit of phycocyanin. We have overproduced CpcE and CpcF in *Escherichia coli*. In vitro, these proteins catalyze the attachment of phycocyanobilin to the .alpha. subunit of apophycocyanin at the appropriate site, .alpha.-Cys-84, to form the correct adduct. CpcE and CpcF also efficiently catalyze the reverse reaction, in which the bilin from holo-.alpha. subunit is transferred either to the apo-.alpha. subunit of the same C-phycocyanin or to the apo-.alpha. subunit of a heterologous C-phycocyanin. The forward and reverse reactions each require both CpcE and CpcF and are specific for the .alpha.-Cys-84 position. Phycocyanobilin is the immediate precursor of the protein-bound bilin.

ACCESSION NUMBER: 92240711 EMBASE
DOCUMENT NUMBER: 1992240711
TITLE: Phycocyanin .alpha.-subunit phycocyanobilin lyase.
AUTHOR: Fairchild C.D.; Zhao J.; Zhou J.; Colson S.E.; Bryant D.A.; Glazer A.N.
CORPORATE SOURCE: MCB: Stanley/Donner ASU, 229 Stanley Hall, University of California, Berkeley, CA 94720, United States
SOURCE: Proceedings of the National Academy of Sciences of the United States of America, (1992) 89/15 (7017-7021).
ISSN: 0027-8424 CODEN: PNASA6
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 029 Clinical Biochemistry
LANGUAGE: English
SUMMARY LANGUAGE: English

=> d his

FILE 'MEDLINE, BIOSIS, EMBASE, DGENE, WPIDS, FSTA, JICST-EPLUS, JAPIO'
ENTERED AT 15:17:25 ON 28 MAR 2003

L1 143 S PHYCOERYTHROCYANIN
L2 9 S HOLO ALPHA SUBUNIT
L3 3 S L2 AND L1

=> d l3 ti abs ibib tot

L3 ANSWER 1 OF 3 MEDLINE

TI Biosynthesis of the cyanobacterial light-harvesting polypeptide
phycoerythrocyanin holo-alpha subunit
in a heterologous host.

AB The entire pathway for the biosynthesis of the phycobiliviolin-bearing His-tagged **holo-alpha subunit** of the cyanobacterial photosynthetic accessory protein **phycoerythrocyanin** was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to 3Z-phycocyanobilin, a precursor of phycobiliviolin (namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase), were expressed from a plasmid under the control of the hybrid trp-lac (trc) promoter. Genes for the apo-**phycoerythrocyanin** alpha subunit (pecA) and the heterodimeric lyase/isomerase (pecE and pecF), which catalyzes both the covalent attachment of phycocyanobilin and its concurrent isomerization to phycobiliviolin, were expressed from the trc promoter on a second plasmid. Upon induction, recombinant *E. coli* used endogenous heme to produce holo-PecA with absorbance and fluorescence properties similar to those of the same protein produced in cyanobacteria. About two-thirds of the apo-PecA was converted to holo-PecA. No significant bilin addition took place in a similarly engineered *E. coli* strain that lacks pecE and pecF. By using immobilized metal affinity chromatography, both apo-PecA and holo-PecA were isolated as ternary complexes with PecE and PecF. The identities of all three components in the ternary complexes were established unambiguously by protein and tryptic peptide analyses performed by matrix-assisted laser desorption ionization-time of flight mass spectrometry.

ACCESSION NUMBER: 2002415572 MEDLINE

DOCUMENT NUMBER: 22159919 PubMed ID: 12169589

TITLE: Biosynthesis of the cyanobacterial light-harvesting polypeptide **phycoerythrocyanin holo-alpha subunit** in a heterologous host.

AUTHOR: Tooley Aaron J; Glazer Alexander N

CORPORATE SOURCE: Department of Molecular and Cell Biology, University of California, Berkeley, CA 94720-3200, USA.

SOURCE: JOURNAL OF BACTERIOLOGY, (2002 Sep) 184 (17) 4666-71.
Journal code: 2985120R. ISSN: 0021-9193.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200208

ENTRY DATE: Entered STN: 20020810

Last Updated on STN: 20020830

Entered Medline: 20020829

L3 ANSWER 2 OF 3 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.

TI Biosynthesis of the cyanobacterial light-harvesting polypeptide
phycoerythrocyanin holo-alpha subunit
in a heterologous host.

AB The entire pathway for the biosynthesis of the phycobiliviolin-bearing His-tagged **holo-alpha subunit** of the cyanobacterial photosynthetic accessory protein **phycoerythrocyanin** was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to 3Z-phycocyanobilin, a

precursor of phycobiliviolin (namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase), were expressed from a plasmid under the control of the hybrid trp-lac (trc) promoter. Genes for the apo-**phycoerythrocyanin** alpha subunit (pecA) and the heterodimeric lyase/isomerase (pecE and pecF), which catalyzes both the covalent attachment of phycocyanobilin and its concurrent isomerization to phycobiliviolin, were expressed from the trc promoter on a second plasmid. Upon induction, recombinant E. coli used endogenous heme to produce holo-PecA with absorbance and fluorescence properties similar to those of the same protein produced in cyanobacteria. About two-thirds of the apo-PecA was converted to holo-PecA. No significant bilin addition took place in a similarly engineered E. coli strain that lacks pecE and pecF. By using immobilized metal affinity chromatography, both apo-PecA and holo-PecA were isolated as ternary complexes with PecE and PecF. The identities of all three components in the ternary complexes were established unambiguously by protein and tryptic peptide analyses performed by matrix-assisted laser desorption ionization-time of flight mass spectrometry.

ACCESSION NUMBER: 2002:464629 BIOSIS
DOCUMENT NUMBER: PREV200200464629
TITLE: Biosynthesis of the cyanobacterial light-harvesting polypeptide **phycoerythrocyanin holo-alpha subunit** in a heterologous host.
AUTHOR(S): Tooley, Aaron J.; Glazer, Alexander N. (1)
CORPORATE SOURCE: (1) Natural Reserve System, University of California, 1111 Franklin Street, 6th Floor, Oakland, CA, 94607-5200:
glazer@uclink4.berkeley.edu USA
SOURCE: Journal of Bacteriology, (September, 2002) Vol. 184, No. 17, pp. 4666-4671. <http://intl-jb.asm.org/>. print.
ISSN: 0021-9193.
DOCUMENT TYPE: Article
LANGUAGE: English

L3 ANSWER 3 OF 3 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.
TI Biosynthesis of the cyanobacterial light-harvesting polypeptide **phycoerythrocyanin holo-.alpha. subunit** in a heterologous host.
AB The entire pathway for the biosynthesis of the phycobiliviolin-bearing His-tagged **holo-.alpha. subunit** of the cyanobacterial photosynthetic accessory protein **phycoerythrocyanin** was reconstituted in Escherichia coli. Cyanobacterial genes encoding enzymes required for the conversion of heme to 3Z-phycocyanobilin, a precursor of phycobiliviolin (namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase), were expressed from a plasmid under the control of the hybrid trp-lac (trc) promoter. Genes for the apo-**phycoerythrocyanin .alpha. subunit** (pecA) and the heterodimeric lyase/isomerase (pecE and pecF), which catalyzes both the covalent attachment of phycocyanobilin and its concurrent isomerization to phycobiliviolin, were expressed from the trc promoter on a second plasmid. Upon induction, recombinant E. coli used endogenous heme to produce holo-PecA with absorbance and fluorescence properties similar to those of the same protein produced in cyanobacteria. About two-thirds of the apo-PecA was converted to holo-PecA. No significant bilin addition took place in a similarly engineered E. coli strain that lacks pecE and pecF. By using immobilized metal affinity chromatography, both apo-PecA and holo-PecA were isolated as ternary complexes with PecE and PecF. The identities of all three components in the ternary complexes were established unambiguously by protein and tryptic peptide analyses performed by matrix-assisted laser desorption ionization-time of flight mass spectrometry.

ACCESSION NUMBER: 2002295599 EMBASE
TITLE: Biosynthesis of the cyanobacterial light-harvesting polypeptide **phycoerythrocyanin holo-.alpha. subunit** in a heterologous host.

AUTHOR: Tooley A.J.; Glazer A.N.
CORPORATE SOURCE: A.N. Glazer, Natural Reserve System, University of California, 1111 Franklin Street, Oakland, CA 94607-5200, United States. glazer@uclink4.berkeley.edu
SOURCE: Journal of Bacteriology, (2002) 184/17 (4666-4671).
Refs: 22
ISSN: 0021-9193 CODEN: JOBAAY
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English

=> d his

(FILE 'HOME' ENTERED AT 15:16:17 ON 28 MAR 2003)

FILE 'MEDLINE, BIOSIS, EMBASE, DGENE, WPIDS, FSTA, JICST-EPLUS, JAPIO'
ENTERED AT 15:17:25 ON 28 MAR 2003

L1 143 S PHYCOERYTHROCYANIN
L2 9 S HOLO ALPHA SUBUNIT
L3 3 S L2 AND L1

=> s phycobiliviolin
L4 21 PHYCOBILIVIOLIN

=> s phycobiliprotein
L5 724 PHYCOBILIPROTEIN

=> s l1 and l5
L6 39 L1 AND L5

=> s l6 and l2
L7 1 L6 AND L2

=> d 17 ti abs ibib tot

L7 ANSWER 1 OF 1 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.
TI Biosynthesis of the cyanobacterial light-harvesting polypeptide phycoerythrocyanin holo-.alpha. subunit in a heterologous host.

AB The entire pathway for the biosynthesis of the phycobiliviolin-bearing His-tagged holo-.alpha. subunit of the cyanobacterial photosynthetic accessory protein phycoerythrocyanin was reconstituted in Escherichia coli. Cyanobacterial genes encoding enzymes required for the conversion of heme to 3Z-phycocyanobilin, a precursor of phycobiliviolin (namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase), were expressed from a plasmid under the control of the hybrid trp-lac (trc) promoter. Genes for the apo-phycoerythrocyanin ..alpha. subunit (pecA) and the heterodimeric lyase/isomerase (pecE and pecF), which catalyzes both the covalent attachment of phycocyanobilin and its concurrent isomerization to phycobiliviolin, were expressed from the trc promoter on a second plasmid. Upon induction, recombinant E. coli used endogenous heme to produce holo-PecA with absorbance and fluorescence properties similar to those of the same protein produced in cyanobacteria. About two-thirds of the apo-PecA was converted to holo-PecA. No significant bilin addition took place in a similarly engineered E. coli strain that lacks pecE and pecF. By using immobilized metal affinity chromatography, both apo-PecA and holo-PecA were isolated as ternary complexes with PecE and PecF. The identities of all three components in the ternary complexes were established unambiguously by protein and tryptic peptide analyses performed by matrix-assisted laser desorption ionization-time of flight

mass spectrometry.
ACCESSION NUMBER: 2002295599 EMBASE
TITLE: Biosynthesis of the cyanobacterial light-harvesting polypeptide phycoerythrocyanin holo-alpha. subunit in a heterologous host.
AUTHOR: Tooley A.J.; Glazer A.N.
CORPORATE SOURCE: A.N. Glazer, Natural Reserve System, University of California, 1111 Franklin Street, Oakland, CA 94607-5200, United States. glazer@uclink4.berkeley.edu
SOURCE: Journal of Bacteriology, (2002) 184/17 (4666-4671).
Refs: 22
ISSN: 0021-9193 CODEN: JOBAAY
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English

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(FILE 'HOME' ENTERED AT 15:16:17 ON 28 MAR 2003)

FILE 'MEDLINE, BIOSIS, EMBASE, DGENE, WPIDS, FSTA, JICST-EPLUS, JAPIO'
ENTERED AT 15:17:25 ON 28 MAR 2003
L1 143 S PHYCOERYTHROCYANIN
L2 9 S HOLO ALPHA SUBUNIT
L3 3 S L2 AND L1
L4 21 S PHYCOPHOBILIVIOLETTIN
L5 724 S PHYCOPHOBILIPROTEIN
L6 39 S L1 AND L5
L7 1 S L6 AND L2

=> d 16 ti abs ibib 1-15

L6 ANSWER 1 OF 39 MEDLINE
TI Novel activity of a phycobiliprotein lyase: both the attachment of phycocyanobilin and the isomerization to phycoviolobilin are catalyzed by the proteins PecE and PecF encoded by the phycoerythrocyanin operon.
AB The structure of phycoviolobilin, the photoactive chromophore of alpha-phycoerythrocyanin, is incompatible with a chromophore ligation to the apoprotein via SH-addition (cysteine) to a Delta3, 3(1)-double bond of the phycobilin. The two putative phycoerythrocyanin lyase genes of *Mastigocladus laminosus*, *pecE* and *pecF*, were overexpressed in *Escherichia coli*. Their action has been studied on the addition reaction of phycocyanobilin to apo-alpha-phycoerythrocyanin (PecA). In the absence of the components of alpha-PEC-phycoviolobilin lyase PecE and PecF, or in the presence of only one of them, phycocyanobilin binds covalently to PecA forming a fluorescent chromoprotein with a red-shifted absorption ($\lambda_{max}=641$ nm) and low photoactivity (<10%). In the presence of both PecE and PecF, a chromoprotein forms which by its absorption ($\lambda_{max}=565$ nm) and high photoreversible photochromism (100% type I) has been identified as integral alpha-phycoerythrocyanin. We conclude that PecE and PecF jointly catalyze not only the addition of phycocyanobilin to PecA, but also its isomerization to the native phycoviolobilin chromophore.

ACCESSION NUMBER: 2000175401 MEDLINE
DOCUMENT NUMBER: 20175401 PubMed ID: 10708746
TITLE: Novel activity of a phycobiliprotein lyase: both the attachment of phycocyanobilin and the isomerization to phycoviolobilin are catalyzed by the proteins PecE and PecF encoded by the phycoerythrocyanin operon.
AUTHOR: Zhao K H; Deng M G; Zheng M; Zhou M; Parbel A; Storf M;

CORPORATE SOURCE: Meyer M; Strohmann B; Scheer H
College of Life Sciences, Wuhan University, Wuhan, PR
China.. khzhao@public.wh.hb.cn

SOURCE: FEBS LETTERS, (2000 Mar 3) 469 (1) 9-13.
Journal code: 0155157. ISSN: 0014-5793.

PUB. COUNTRY: Netherlands

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200004

ENTRY DATE: Entered STN: 20000421
Last Updated on STN: 20000421
Entered Medline: 20000410

L6 ANSWER 2 OF 39 MEDLINE

TI A role for cpeYZ in cyanobacterial phycoerythrin biosynthesis.

AB Pigment mutant strain FdR1 of the filamentous cyanobacterium *Fremyella diplosiphon* is characterized by constitutive synthesis of the **phycobiliprotein** phycoerythrin due to insertional inactivation of the rcaC regulatory gene by endogenous transposon Tn5469. Whereas the parental strain Fd33 harbors five genomic copies of Tn5469, cells of strain FdR1 harbor six genomic copies of the element; the sixth copy in FdR1 is localized to the rcaC gene. Electroporation of FdR1 cells yielded secondary pigment mutant strains FdR1E1 and FdR1E4, which identically exhibited the FdR1 phenotype with significantly reduced levels of phycoerythrin. In both FdR1E1 and FdR1E4, a seventh genomic copy of Tn5469 was localized to the cpeY gene of the sequenced but phenotypically uncharacterized cpeYZ gene set. This gene set is located downstream of the cpeBA operon which encodes the alpha and beta subunits of phycoerythrin. Complementation experiments correlated cpeYZ activity to the phenotype of strains FdR1E1 and FdR1E4. The predicted CpeY and CpeZ proteins share significant sequence identity with the products of homologous cpeY and cpeZ genes reported for *Pseudanabaena* sp. strain PCC 7409 and *Synechococcus* sp. strain WH 8020, both of which synthesize phycoerythrin. The CpeY and CpeZ proteins belong to a family of structurally related cyanobacterial proteins that includes the subunits of the CpcE/CpcF phycocyanin alpha-subunit lyase of *Synechococcus* sp. strain PCC 7002 and the subunits of the PecE/PecF **phycoerythrocyanin** alpha-subunit lyase of *Anabaena* sp. strain PCC 7120. Phycobilisomes isolated from mutant strains FdR1E1 and FdR1E4 contained equal amounts of chromophorylated alpha and beta subunits of phycoerythrin at 46% of the levels of the parental strain FdR1. These results suggest that the cpeYZ gene products function in phycoerythrin synthesis, possibly as a lyase involved in the attachment of phycoerythrobilin to the alpha or beta subunit.

ACCESSION NUMBER: 97175521 MEDLINE

DOCUMENT NUMBER: 97175521 PubMed ID: 9023176

TITLE: A role for cpeYZ in cyanobacterial phycoerythrin biosynthesis.

AUTHOR: Kahn K; Mazel D; Houmar J; Tandeau de Marsac N; Schaefer M R

CORPORATE SOURCE: School of Biological Sciences, University of Missouri-Kansas City, 64110, USA.

SOURCE: JOURNAL OF BACTERIOLOGY, (1997 Feb) 179 (4) 998-1006.
Journal code: 2985120R. ISSN: 0021-9193.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

OTHER SOURCE: GENBANK-X04592

ENTRY MONTH: 199703

ENTRY DATE: Entered STN: 19970321
Last Updated on STN: 19990129
Entered Medline: 19970307

L6 ANSWER 3 OF 39 MEDLINE
TI Candidate genes for the **phycoerythrocyanin** alpha subunit lyase.
Biochemical analysis of pecE and pecF interposon mutants.
AB The rod substructures of the Anabaena sp. PCC 7120 phycobilisome contain the light harvesting proteins C-phycocyanin and **phycoerythrocyanin** (PEC). Even at low light intensities, PEC represents no more than 5% of the phycobilisome protein. The beta subunits of both proteins carry thioether-linked phycocyanobilin (PCB) at beta-Cys-82 and beta-Cys-155; however, C-phycocyanin has PCB at alpha-Cys-84 whereas PEC alpha subunit carries phycobiliviolin at this position. The Anabaena sp. PCC 7120 pec operon is made up of five genes. PecB and pecA encode the beta and alpha subunits of PEC, pecC encodes a linker polypeptide associated with PEC in the rod substructure, and pecE and pecF are genes of unknown function that show a high degree of homology to cpcE and cpcF, that encode a C-phycocyanin alpha subunit PCB lyase (Fairchild, C. D., Zhao, J., Zhou, J., Colson, S. E., Bryant, D. A., and Glazer, A. N. (1992) Proc. Natl. Acad. Sci. U.S.A. 89, 7017-7021). Insertional mutants in pecE and pecF, and an interposon mutant in which a portion of both pecE and pecF was deleted, were constructed. All three types of mutants grew 1.3 times slower than wild-type under limiting light conditions and showed a 20% reduction in the PCB content of whole cells relative to chlorophyll alpha. Holo-PEC was missing from the phycobilisomes of all three types of mutants and the level of the PEC linker polypeptide was reduced relative to the wild-type. However, approximately 30% of the wild-type level of the PEC beta subunit was present in all of these phycobilisomes. In contrast, the PEC alpha subunit was barely detectable in the pecE and pecF mutants, but was present in the pecEF deletion mutant as a PCB-adduct in a 1:1 ratio with the PEC beta subunit. The identity of this "unnatural" adduct was confirmed by isolation of the subunit and amino-terminal sequencing. These biochemical results support the inference that pecE and pecF encode a PEC alpha subunit phycobiliviolin lyase, and, in conjunction with earlier findings, demonstrate that **phycobiliprotein** bilin lyases show high selectivity (rather than absolute specificity) for both the bilin and the polypeptide substrate.

ACCESSION NUMBER: 95279433 MEDLINE
DOCUMENT NUMBER: 95279433 PubMed ID: 7759546
TITLE: Candidate genes for the **phycoerythrocyanin** alpha subunit lyase. Biochemical analysis of pecE and pecF interposon mutants.
AUTHOR: Jung L J; Chan C F; Glazer A N
CORPORATE SOURCE: Department of Molecular and Cell Biology, University of California, Berkeley 94720, USA.
CONTRACT NUMBER: GM28994 (NIGMS)
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (1995 May 26) 270 (21) 12877-84.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199506
ENTRY DATE: Entered STN: 19950707
Last Updated on STN: 19950707
Entered Medline: 19950628

L6 ANSWER 4 OF 39 MEDLINE
TI The complete amino acid sequence of R-phycocyanin-I alpha and beta subunits from the red alga Porphyridium cruentum. Structural and phylogenetic relationships of the phycocyanins within the **phycobiliprotein** families.
AB We present here the complete primary structure of R-phycocyanin-I alpha and beta subunits from the red alga Porphyridium cruentum. The alpha chain is composed of 162 amino acid residues (18049 Da, calculated from sequence, including chromophore) and carries a phycocyanobilin pigment

covalently linked to Cys84. The beta chain contains 172 amino acids (19344Da, calculated from sequence, including chromophores) and carries a phycocyanobilin pigment covalently linked at Cys82 and a phycoerythrobilin pigment at Cys153. A gamma-N-methyl asparagine residue was also characterised at position beta 72 similar to other **phycobiliprotein** beta subunits. R-phycocyanin-I from *Porphyridium cruentum* shares high sequence identity with C-phycocyanins (69-83%), R-phycocyanins (66-70%) and in a less extent with phycoerythrocyanins (57-65%) from various sources. The presented phylogenetic trees are based on a comparison of all **phycobiliprotein** amino acid sequences known so far and confirm the clear affiliation of the R-phycocyanins in the phycocyanin family. In spite of their particular phycobilin pattern, they do not represent intermediate forms between the phycocyanin and the phycoerythrin family. **Phycoerythrocyanin**, a phycocyanin-related **phycobiliprotein** adapted to green light harvesting, is also shown to belong to the phycocyanin family. However, the phycoerythrocyanins diverge from phycocyanins in their different function and it is suggested that they should be assigned to a separate group within the phycocyanin family.

ACCESSION NUMBER: 94222105 MEDLINE
DOCUMENT NUMBER: 94222105 PubMed ID: 8168545
TITLE: The complete amino acid sequence of R-phycocyanin-I alpha and beta subunits from the red alga *Porphyridium cruentum*. Structural and phylogenetic relationships of the phycocyanins within the **phycobiliprotein** families.
AUTHOR: Ducret A; Sidler W; Frank G; Zuber H
CORPORATE SOURCE: Institute for Molecular Biology and Biophysics, Federal Institute of Technology, Zurich, Switzerland.
SOURCE: EUROPEAN JOURNAL OF BIOCHEMISTRY, (1994 Apr 1) 221 (1) 563-80.
PUB. COUNTRY: Journal code: 0107600. ISSN: 0014-2956.
DOCUMENT TYPE: GERMANY: Germany, Federal Republic of
LANGUAGE: Journal; Article; (JOURNAL ARTICLE)
FILE SEGMENT: English
ENTRY MONTH: Priority Journals
199406
ENTRY DATE: Entered STN: 19940613
Last Updated on STN: 19940613
Entered Medline: 19940602

L6 ANSWER 5 OF 39 MEDLINE
TI Structure of the genes encoding the rod-core linker polypeptides of *Mastigocladus laminosus* phycobilisomes and functional aspects of the **phycobiliprotein**/linker-polypeptide interactions.
AB The 3' portion of the cpc operon in *Mastigocladus laminosus* encloses the genes 5'-cpcF-cpcG1-cpcG2-cpcG3 3'. The three cpcG genes encode different phycocyanin-associated rod-core linker polypeptides of the phycobilisomes with predicted 279, 247 and 254 amino acids in length. The gene products CpcG show a high similarity at their N-terminal domains (190 amino acids) and an overall identity of 47-53% to one another. Each of the three CpcG polypeptides is highly related to one of the four CpcG gene products of *Anabaena* sp. PCC 7120 (66-81% identity). It is suggested that these pairs of rod-core linker polypeptides mediate the same specific type of phycocyanin---allophycocyanin interaction in the similar phycobilisomes of *M. laminosus* and *Anabaena* sp. PCC 7120. The similarity of the CpcG1, CpcG2 and CpcG3 polypeptides to the single CpcG rod-core linker polypeptide of *Synechococcus* sp. PCC 7002 (36-41% identity) is lower. The rod-core linker polypeptides are more distantly related to the rod linker polypeptides associated with phycocyanin or phycoerythrin. However, six conserved domains were identified within the N-terminal 190 amino acids of these linker proteins, which bear similar amino acid sequences, including highly conserved basic amino acids. A similar amino acid sequence but with conserved acidic amino acids can be found in the beta subunits of

phycocyanin, phycoerythrin and **phycoerythrocyanin**, which is protruding into the central cavity of the **phycobiliprotein hexamers**. It is suggested that these domains are sites of **phycobiliprotein-hexamer/rod** and **rod-core linker interactions**.

ACCESSION NUMBER: 92249337 MEDLINE
DOCUMENT NUMBER: 92249337 PubMed ID: 1577010
TITLE: Structure of the genes encoding the rod-core linker polypeptides of *Mastigocladus laminosus* phycobilisomes and functional aspects of the **phycobiliprotein** /linker-polypeptide interactions.
AUTHOR: Glauser M; Stirewalt V L; Bryant D A; Sidler W; Zuber H
CORPORATE SOURCE: Institut fur Molekularbiologie und Biophysik,
Eidgenossische Technische Hochschule, Zurich, Switzerland.
CONTRACT NUMBER: GM-31625 (NIGMS)
SOURCE: EUROPEAN JOURNAL OF BIOCHEMISTRY, (1992 May 1) 205 (3)
927-37.
Journal code: 0107600. ISSN: 0014-2956.
PUB. COUNTRY: GERMANY: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-S87180; GENBANK-S87218; GENBANK-S87221;
GENBANK-S87223; GENBANK-X59763; GENBANK-X64458;
GENBANK-X65178; GENBANK-X65179; GENBANK-X65180;
GENBANK-X65181
ENTRY MONTH: 199206
ENTRY DATE: Entered STN: 19920619
Last Updated on STN: 19920619
Entered Medline: 19920609

L6 ANSWER 6 OF 39 MEDLINE
TI Refined three-dimensional structure of **phycoerythrocyanin** from the cyanobacterium *Mastigocladus laminosus* at 2.7 Å.
AB The structure of the **phycobiliprotein phycoerythrocyanin** from the thermophilic cyanobacterium *Mastigocladus laminosus* has been determined at 2.7 Å resolution by X-ray diffraction methods on the basis of the molecular model of C-phycocyanin from the same organism. Hexagonal **phycoerythrocyanin** crystals of space group P6(3) with cell constants $a = b = 156.86 \text{ Å}$, $c = 40.39 \text{ Å}$, $\alpha = \beta = 90$ degrees, $\gamma = 120$ degrees are almost isomorphous to C-phycocyanin crystals. The crystal structure has been refined by energy-restrained crystallographic refinement and model building. The conventional crystallographic R-factor of the final model was 19.2% with data to 2.7 Å resolution. In **phycoerythrocyanin**, the three (α β)-subunits are arranged around a 3-fold symmetry axis, as in C-phycocyanin. The two structures are very similar. After superposition, the 162 C α atoms of the α -subunit have a mean difference of 0.71 Å and the 171 C α atoms of the β -subunit differ by 0.51 Å. The stereochemistry of the chiral atoms in the phycobiliviolin chromophore A84 is C(31)-R, C(4)-S. The configuration of the chromophore is C(10)-Z, C(15)-Z and the conformation C(5)-anti, C(9)-syn and C(14)-anti like the phycocyanobilin chromophores in **phycoerythrocyanin** and C-phycocyanin.

ACCESSION NUMBER: 90172426 MEDLINE
DOCUMENT NUMBER: 90172426 PubMed ID: 2106585
TITLE: Refined three-dimensional structure of **phycoerythrocyanin** from the cyanobacterium *Mastigocladus laminosus* at 2.7 Å.
AUTHOR: Duerring M; Huber R; Bode W; Ruembeli R; Zuber H
CORPORATE SOURCE: Max-Planck Institut fur Biochemie, Martinsried, Germany.
SOURCE: JOURNAL OF MOLECULAR BIOLOGY, (1990 Feb 5) 211 (3) 633-44.
Journal code: 2985088R. ISSN: 0022-2836.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English

FILE SEGMENT: Priority Journals
ENTRY MONTH: 199004
ENTRY DATE: Entered STN: 19900601
Last Updated on STN: 19900601
Entered Medline: 19900409

L6 ANSWER 7 OF 39 MEDLINE
TI Crosslinking of phycobiliproteins from the cyanobacterium *Mastigocladus laminosus* with bis-imidates: localization of an intrasubunit and an intersubunit crosslink in C-phycocyanin.
AB The light-harvesting pigment-protein complexes allophycocyanin (AP), C-phycocyanin (PC) and phycoerythrocyanin (PEC) of the cyanobacterium *Mastigocladus laminosus* consist of alpha- and beta-subunits containing about 170 amino-acid residues each. These two subunits form an alpha,beta-monomer, three of which build up a disc-shaped trimer. In this study these phycobiliproteins were crosslinked with bis-imidates. Various spacer lengths of the reagent and various aggregation states of the phycobiliprotein were tested. An intersubunit crosslink could be verified in all three phycobiliproteins. PC-trimmers were crosslinked with the homobifunctional reagent dimethyl pimelimidate having a maximal crosslinking distance of 10 Å. Two crosslinks could be identified: an intramonomer intersubunit crosslink with a yield of 48% and an intrasubunit crosslink within alpha PC (57%). These products were chemically and enzymatically fragmented and the small crosslinked peptides were isolated and then identified by amino-acid analysis. The following amino acids were crosslinked: alpha-Val 1 with beta-Ala 1 and alpha-Lys 62 with alpha-Lys 134. Both crosslinks could be localized within the known three-dimensional structure of PC.

ACCESSION NUMBER: 88050102 MEDLINE
DOCUMENT NUMBER: 88050102 PubMed ID: 3118901
TITLE: Crosslinking of phycobiliproteins from the cyanobacterium *Mastigocladus laminosus* with bis-imidates: localization of an intrasubunit and an intersubunit crosslink in C-phycocyanin.
AUTHOR: Rumbeli R; Wirth M; Zuber H
CORPORATE SOURCE: Institut fur Molekularbiologie und Biophysik,
Eidgenossische Technische Hochschule, Zurich.
SOURCE: BIOLOGICAL CHEMISTRY HOPPE-SEYLER, (1987 Sep) 368 (9)
1179-91.
Journal code: 8503054. ISSN: 0177-3593.
PUB. COUNTRY: GERMANY, WEST: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198801
ENTRY DATE: Entered STN: 19900305
Last Updated on STN: 19900305
Entered Medline: 19880115

L6 ANSWER 8 OF 39 MEDLINE
TI Linker polypeptides of the phycobilisome from the cyanobacterium *Mastigocladus laminosus*. II. Amino-acid sequences and functions.
AB The complete primary structure of an 80-residue linker polypeptide, LR(C)8.9, from the phycobilisome of the cyanobacterium *Mastigocladus laminosus* was determined as well as the 44 N-terminal residues of the two linker polypeptides LR34.5, PEC and LR34.5, PC and the 114 C-terminal residues of LR34.5, PEC. A brief description of the structure determination and an extensive discussion of the relationships of these polypeptides have been published recently (Fuglistaller, P., Suter, F. & Zuber, H. (1985) Biol. Chem. Hoppe-Seyler 366, 993-1001). In this paper we report in detail about the elucidation of the primary structures. Limited digestion of the hexameric phycobiliprotein-linker polypeptide complex (alpha PEC beta PEC)6LR34.5, PEC with various proteases resulted in a linker polypeptide diminished by a 1-5 kDa segment, while the

phycobiliproteins remained intact. By N-terminal sequence analysis of the residual part of the linker polypeptide in the complex, LR34.5-5, PEC, it was concluded that the C-terminus of the polypeptide had been attacked by the proteases. This C-terminal part of the protein influences the hexamer formation of phycoerythrocyanin (PEC) and is responsible for the linkage between two phycobiliprotein hexamers. From the function of the C-terminal segment of LR34.5, PEC and its homology to the LR(C)8.9 polypeptide, it was concluded that LR(C)8.9 is located at the end of the peripheral phycobilisomal rods distal to the allophycocyanin core.

ACCESSION NUMBER: 87000168 MEDLINE
DOCUMENT NUMBER: 87000168 PubMed ID: 3092841
TITLE: Linker polypeptides of the phycobilisome from the cyanobacterium Mastigocladus laminosus. II. Amino-acid sequences and functions.
AUTHOR: Fuglistaller P; Suter F; Zuber H
SOURCE: BIOLOGICAL CHEMISTRY HOPPE-SEYLER, (1986 Jul) 367 (7) 615-26.
Journal code: 8503054. ISSN: 0177-3593.
PUB. COUNTRY: GERMANY, WEST: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198611
ENTRY DATE: Entered STN: 19900302
Last Updated on STN: 19900302
Entered Medline: 19861114

L6 ANSWER 9 OF 39 MEDLINE

TI Linker polypeptides of the phycobilisome from the cyanobacterium Mastigocladus laminosus. I. Isolation and characterization of phycobiliprotein-linker-polypeptide complexes.
AB Phycobilisomes from the cyanobacterium Mastigocladus laminosus cultured in white and red light were isolated and compared with respect to the phycoerythrocyanin (PEC) and linker polypeptide contents. It was verified that the production of PEC is induced by low light intensities. A PEC complex, (alpha PEC beta PEC)6LR34.5, PEC, and a phycocyanin (PC) complex, (alpha PC beta PC)6LR34.5, PC, were isolated from phycobilisomes by Cellex-D anion exchange chromatography and sucrose density gradient centrifugation. The absorption and fluorescence emission maxima of the PEC complex are at 575 and 620 nm and those of the PC complex are at 631 and 647 nm, respectively. The extinction coefficients of the two complexes were determined. From different experiments it was concluded that PEC is present as a hexameric complex, (alpha PEC beta PEC)6LR34.5, PEC, in the phycobilisome. The two linker polypeptides LR34.5, PEC and LR34.5, PC were isolated from their phycobiliprotein complexes by gel filtration on Bio-Gel P-100 in 50% formic acid. A 5-kDa terminal segment of both linker polypeptides was found to influence the hexamer formation of the phycobiliproteins. The same segments have been described to be responsible for the hexamer-hexamer linkage (Yu, M.-H. & Glazer, A.N. (1982) J. Biol. Chem. 257, 3429-3433). A 8.9-kDa linker polypeptide, LR(C)8.9, was isolated from a PEC fraction of the Cellex-D column by Bio-Gel P-100 gel filtration in 50% formic acid. Localisation of this protein within the phycobilisome was attempted. Its most probable function is to terminate the phycobilisomal rods at the end distal to the allophycocyanin core.

ACCESSION NUMBER: 87000167 MEDLINE
DOCUMENT NUMBER: 87000167 PubMed ID: 3092840
TITLE: Linker polypeptides of the phycobilisome from the cyanobacterium Mastigocladus laminosus. I. Isolation and characterization of phycobiliprotein-linker-polypeptide complexes.
AUTHOR: Fuglistaller P; Suter F; Zuber H
SOURCE: BIOLOGICAL CHEMISTRY HOPPE-SEYLER, (1986 Jul) 367 (7) 601-14.
Journal code: 8503054. ISSN: 0177-3593.

PUB. COUNTRY: GERMANY, WEST: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198611
ENTRY DATE: Entered STN: 19900302
Last Updated on STN: 19900302
Entered Medline: 19861114

L6 ANSWER 10 OF 39 MEDLINE
TI Linker polypeptides of the phycobilisome from the cyanobacterium
Mastigocladus laminosus: amino-acid sequences and relationships.
AB Three linker polypeptides of the phycobilisome from the cyanobacterium
Mastigocladus laminosus were isolated: A 8.9-kDa polypeptide, L8.9R(C),
which is probably associated with C-phycocyanin, a 34.5-kDa polypeptide,
L34.5,PCR, which forms a complex with C-phycocyanin, and a 34.5-kDa
polypeptide, L34.5,PECR, which is linked to **phycoerythrocyanin**.
The complete amino-acid sequence (80 residues) of the L8.9R(C) polypeptide
was determined as well as the N-terminal 44 residues of both L34.5R
polypeptides and the 114 C-terminal residues of L34.5,PECR. L8.9R(C) is
homologous to L8.9C (Fuglistaller et al. (1984) Hoppe-Seyler's Z. Physiol.
Chem. 365, 1085-1096) and to the C-terminal sequence of L34.5,PECR. The
N-terminal sequences of L34.5,PECR and L34.5,PCR exhibit 34% homology. The
44 N-terminal residues of L34.5,PECR are related to the beta-subunit of
phycoerythrocyanin (23% homology), while the C-terminal sequence
of L34.5,PECR is more related to alpha PEC (21% homology within 60
residues). This suggests that the 30-kDa-linker polypeptide family
originates from a fusion of the alpha- and beta-subunit genes and the
corresponding intercistronic DNA sequence, as might have arisen through
mutation in the stop-codon of the beta-subunit gene. Hence, all
polypeptides of the phycobilisome (including perhaps the anchor
polypeptide) may be derived from an early ancestor
phycobiliprotein subunit, which itself is also related to
myoglobin (Schirmer et al. (1985) J. Mol. Biol. 184, 251-277).
ACCESSION NUMBER: 86050914 MEDLINE
DOCUMENT NUMBER: 86050914 PubMed ID: 3933528
TITLE: Linker polypeptides of the phycobilisome from the
cyanobacterium Mastigocladus laminosus: amino-acid
sequences and relationships.
AUTHOR: Fuglistaller P; Suter F; Zuber H
SOURCE: BIOLOGICAL CHEMISTRY HOPPE-SEYLER, (1985 Oct) 366 (10)
993-1001.
Journal code: 8503054. ISSN: 0177-3593.
PUB. COUNTRY: GERMANY, WEST: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198601
ENTRY DATE: Entered STN: 19900321
Last Updated on STN: 19900321
Entered Medline: 19860117

L6 ANSWER 11 OF 39 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
TI Characterization of phycoviolobilin **phycoerythrocyanin**
-alpha84-cystein-lyase-(isomerizing) from Mastigocladus laminosus.
AB Cofactor requirements and enzyme kinetics have been studied of the novel,
dual-action enzyme, the isomerizing phycoviolobilin
phycoerythrocyanin-alpha84-cystein-lyase (PVB-PEC-lyase) from
Mastigocladus laminosus, which catalyses both the covalent attachment of
phycocyanobilin to PecA, the apo-alpha-subunit of
phycoerythrocyanin, and its isomerization to phycoviolobilin.
Thiols and the divalent metals, Mg²⁺ or Mn²⁺, were required, and the
reaction was aided by the detergent, Triton X-100. Phosphate buffer
inhibits precipitation of the proteins present in the reconstitution

mixture, but at the same time binds the required metal. Kinetic constants were obtained for both substrates, the chromophore ($K_m=12-16 \mu M$, depending on (PecA), $k_{cat}apprx eq1.2\times 10^{-4} \text{cntdots-1}$) and the apoprotein ($K_m=2.4 \mu M$ at $14 \mu M$ PCB, $k_{cat}=0.8\times 10^{-4} \text{cntdots-1}$). The kinetic analysis indicated that the reconstitution reaction proceeds by a sequential mechanism. By a combination of untagged and His-tagged subunits, evidence was obtained for a complex formation between PecE and PecF (subunits of PVB-PEC-lyase), and by experiments with single subunits for the prevalent function of PecE in binding and PecF in isomerizing the chromophore.

ACCESSION NUMBER: 2002:561644 BIOSIS

DOCUMENT NUMBER: PREV200200561644

TITLE: Characterization of phycobilobilin
phycoerythrocyanin-alpha84-cystein-lyase-
(isomerizing) from Mastigocladus laminosus.

AUTHOR(S): Zhao, Kai-Hong (1); Wu, Dong; Wang, Lu; Zhou, Ming; Storf, Max; Bubenzer, Claudia; Strohmann, Brigitte; Scheer, Hugo

CORPORATE SOURCE: (1) College of Life Science and Technology, Huazhong University of Science and Technology, Wuhan, Hubei, 430074:
khzhao@163.com, scheer-h@botanik.biologie.uni-muenchen.de
China

SOURCE: European Journal of Biochemistry, (September, 2002) Vol. 269, No. 18, pp. 4542-4550. <http://www.blackwell-science.com/cgilib/jnlpage.asp?journal=ejb&file=ejb&page=a>
ims. print.

ISSN: 0014-2956.

DOCUMENT TYPE: Article

LANGUAGE: English

L6 ANSWER 12 OF 39 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.

TI One century of protein crystallography: The phycobiliproteins.

AB The physical principles that underlay the rapid and efficient energy transfer from the light absorbing phycobilisomes to the reaction centre are conceivable from the knowledge of the exact three-dimensional structure of the phycobiliproteins and chromophores that are involved. The structure of the components and their assembly in the phycobilisomes could be determined by the structure analysis of X-ray data derived from phycobiliprotein crystals. Reports about these very aesthetic and brilliantly colored crystals have been published for more than a hundred years but it was only in the last decade that the structures of the different members of the phycobiliprotein family were solved for the first time at atomic resolution - all of them in Martinsried at the Max-Planck-Institut fur Biochemie. Despite the appearance of common structural principles the most important finding was that very subtle modifications in the structure and environment of the chromophores are sufficient to establish a highly specific light harvesting system in which the phycobiliproteins function with great cooperativity and efficiency.

ACCESSION NUMBER: 1997:290832 BIOSIS

DOCUMENT NUMBER: PREV199799590035

TITLE: One century of protein crystallography: The
phycobiliproteins.

AUTHOR(S): Betz, Michael

CORPORATE SOURCE: Max-Planck-Institut fuer Biochemie, Abteilung
Strukturforschung, Am Klopferspitz 18a, D-82152 Martinsried
Germany

SOURCE: Biological Chemistry, (1997) Vol. 378, No. 3-4, pp.
167-176.

ISSN: 1431-6730.

DOCUMENT TYPE: General Review

LANGUAGE: English

L6 ANSWER 13 OF 39 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.

TI A role for cpeYZ in cyanobacterial phycoerythrin biosynthesis.

AB Pigment mutant strain FdR1 of the filamentous cyanobacterium Fremyella diplosiphon is characterized by constitutive synthesis of the

phycobiliprotein phycoerythrin due to insertional inactivation of the rcaC regulatory gene by endogenous transposon Tn5469. Whereas the parental strain Fd33 harbors five genomic copies of Tn5469, cells of strain FdR1 harbor six genomic copies of the element; the sixth copy in FdR1 is localized to the rcaC gene. Electroporation of FdR1 cells yielded secondary pigment mutant strains FdR1E1 and FdR1E4, which identically exhibited the FdR1 phenotype with significantly reduced levels of phycoerythrin. In both FdR1E1 and FdR1E4, a seventh genomic copy of Tn5469 was localized to the cpeY gene of the sequenced but phenotypically uncharacterized cpeYZ gene set. This gene set is located downstream of the cpeBA operon which encodes the alpha and beta subunits of phycoerythrin. Complementation experiments correlated cpeYZ activity to the phenotype of strains FdR1E1 and FdR1E4. The predicted CpeY and CpeZ proteins share significant sequence identity with the products of homologous cpeY and cpeZ genes reported for *Pseudanabaena* sp. strain PCC 7409 and *Synechococcus* sp. strain WH 8020, both of which synthesize phycoerythrin. The CpeY and CpeZ proteins belong to a family of structurally related cyanobacterial proteins that includes the subunits of the CpcE/CpcF phycocyanin alpha-subunit lyase of *Synechococcus* sp. strain PCC 7002 and the subunits of the PecE/PecF **phycoerythrocyanin** alpha-subunit lyase of *Anabaena* sp. strain PCC 7120. Phycobilisomes isolated from mutant strains FdR1E1 and FdR1E4 contained equal amounts of chromophorylated alpha and beta subunits of phycoerythrin at 46% of the levels of the parental strain FdR1. These results suggest that the cpeYZ gene products function in phycoerythrin synthesis, possibly as a lyase involved in the attachment of phycoerythrobilin to the alpha or beta subunit.

ACCESSION NUMBER: 1997:155769 BIOSIS
DOCUMENT NUMBER: PREV199799454972
TITLE: A role for cpeYZ in cyanobacterial phycoerythrin biosynthesis.
AUTHOR(S): Kahn, Katherine; Mazel, Didier; Houmar, Jean; De Marsac, Nicole Tandeau; Schaefer, Michael R. (1)
CORPORATE SOURCE: (1) Univ. Missouri-Kansas City, Sch. Biol. Sci., 5100 Rockhill Rd., Kansas City, MO 64110 USA
SOURCE: Journal of Bacteriology, (1997) Vol. 179, No. 4, pp. 998-1006.
ISSN: 0021-9193.
DOCUMENT TYPE: Article
LANGUAGE: English

L6 ANSWER 14 OF 39 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
TI Candidate genes for the **phycoerythrocyanin** alpha subunit lyase: Biochemical analysis of pecE and pecF interposon mutants.
AB The rod substructures of the *Anabaena* sp. PCC 7120 phycobilisome contain the light harvesting proteins C-phycocyanin and **phycoerythrocyanin** (PEC). Even at low light intensities, PEC represents no more than 5% of the phycobilisome protein. The beta subunits of both proteins carry thioether-linked phycocyanobilin (PCB) at beta-Cys-82 and beta-Cys-155; however, C-phycocyanin has PCB at alpha-Cys-84 whereas PEC a subunit carries phycobiliviolin at this position. The *Anabaena* sp. PCC 7120 pec operon is made up of five genes. PecB and pecA encode the beta and alpha subunits of PEC, pecC encodes a linker polypeptide associated with PEC in the rod substructure, and pecE and pecF are genes of unknown function that show a high degree of homology to cpcE and cpcF, that encode a C-phycocyanin a subunit PCB lyase (Fairchild, C. D., Zhao, J., Zhou, J., Colson, S. E., Bryant, D. A., and Glazer, A. N. (1992) Proc. Natl. Acad. Sci. U. S. A. 89, 7017-7021). Insertional mutants in pecE and pecF, and an interposon mutant in which a portion of both pecE and pecF was deleted, were constructed. All three types of mutants grew 1.3 times slower than wild-type under limiting light conditions and showed a 20% reduction in the PCB content of whole cells relative to chlorophyll alpha. Holo-PEC was missing from the phycobilisomes of all three types of mutants and the level of the PEC linker polypeptide was reduced relative to the wild-type. However, apprx 30% of the wild-type level of the PEC beta subunit was

present in all of these phycobilisomes. In contrast, the PEC α subunit was barely detectable in the *pecE* and *pecF* mutants, but was present in the *pecEF* deletion mutant as a PCB-adduct in a 1:1 ratio with the PEC beta subunit. The identity of this "unnatural" adduct was confirmed by isolation of the subunit and amino-terminal sequencing. These biochemical results support the inference that *pecE* and *pecF* encode a PEC alpha subunit phycobiliviolin lyase, and, in conjunction with earlier findings, demonstrate that **phycobiliprotein** bilin lyases show high selectivity (rather than absolute specificity) for both the bilin and the polypeptide substrate.

ACCESSION NUMBER: 1995:320261 BIOSIS

DOCUMENT NUMBER: PREV199598334561

TITLE: Candidate genes for the **phycoerythrocyanin** alpha subunit lyase: Biochemical analysis of *pecE* and *pecF* interposon mutants.

AUTHOR(S): Jung, Linda J.; Chan, Crystal F.; Glazer, Alexander N. (1)

CORPORATE SOURCE: (1) MCB: Stanley/Donner ASU, 229 Stanley Hall 3206, Univ. Calif., Berkeley, CA 94720-3206 USA

SOURCE: Journal of Biological Chemistry, (1995) Vol. 270, No. 21, pp. 12877-12884.

ISSN: 0021-9258.

DOCUMENT TYPE: Article

LANGUAGE: English

L6 ANSWER 15 OF 39 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.

TI Type I and type II reversible photochemistry of **phycoerythrocyanin** alpha-subunit from *Mastigocladus laminosus* both involve Z, E isomerization of phycobilobilin chromophore and are controlled by sulfhydryls in apoprotein.

AB The alpha-subunit of **phycoerythrocyanin** (α -PEC) can exist in four states (Z-alpha-I, Z-alpha-II, E-alpha-I, E-alpha-II). They are connected pairwise by photoreversible photochromism. The type I photochemistry connecting Z-alpha-I and E-alpha-I, involves a 15Z/E phototransformation. α -PEC showing this type of photochemistry is obtained when the subunits of PEC are separated by gel permeation chromatography in the presence of 63 mM formic acid, or by reduction of the alpha-subunit of **phycoerythrocyanin** of type II reversible photochemistry with mercaptoethanol. α -PEC showing the recently characterized (Hong et al. (1993) Photochem. Photobiol. 58, 745-747) type II photochemistry connecting Z-alpha-II and E-alpha-II can be obtained when the alpha-subunit of **phycoerythrocyanin** of type I photochemistry is allowed to oxidize, or when it is treated with p-chloromercuribenzenesulfonate. The two types of reversible photochemistry of alpha-subunit of phycocerythrocyanin are therefore controlled by the state of the two sulfhydryl group(s), viz. Cys-98,99 of the apoprotein. A quantitative analysis of the PCMS titration showed that modification of either one of these two cysteine residues is sufficient to inhibit type I photochemistry and induces type II. By treatment with mercaptoethanol or PCMS, the end products of type I and type II photochemistry, respectively, could be pairwise transformed into each other, showing that type II also involves 15Z/E isomerization. The difference between them must be due to different interactions between phycobilobilin and apoprotein, which can be modulated by the two sulfhydryls.

ACCESSION NUMBER: 1995:226654 BIOSIS

DOCUMENT NUMBER: PREV199598240954

TITLE: Type I and type II reversible photochemistry of **phycoerythrocyanin** alpha-subunit from *Mastigocladus laminosus* both involve Z, E isomerization of phycobilobilin chromophore and are controlled by sulfhydryls in apoprotein.

AUTHOR(S): Zhao, Kai-Hong; Scheer, Hugo (1)

CORPORATE SOURCE: (1) Botanisches Inst. Univ., Menzinger Str. 67, D-80623, Muenchen Germany

SOURCE: Biochimica et Biophysica Acta, (1995) Vol. 1228, No. 2-3,
pp. 244-253.
ISSN: 0006-3002.

DOCUMENT TYPE: Article
LANGUAGE: English

=> d his

(FILE 'HOME' ENTERED AT 15:16:17 ON 28 MAR 2003)

FILE 'MEDLINE, BIOSIS, EMBASE, DGENE, WPIDS, FSTA, JICST-EPLUS, JAPIO'
ENTERED AT 15:17:25 ON 28 MAR 2003

L1 143 S PHYCOERYTHROCYANIN
L2 9 S HOLO ALPHA SUBUNIT
L3 3 S L2 AND L1
L4 21 S PHYCOBILIVIOLIN
L5 724 S PHYCOBILIPROTEIN
L6 39 S L1 AND L5
L7 1 S L6 AND L2

=> d 16 ti abs ibib 30-39

L6 ANSWER 30 OF 39 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.
TI Biosynthesis of the cyanobacterial light-harvesting polypeptide
phycoerythrocyanin holo-.alpha. subunit in a heterologous host.
AB The entire pathway for the biosynthesis of the phycobiliviolin-bearing
His-tagged holo-.alpha. subunit of the cyanobacterial photosynthetic
accessory protein *phycoerythrocyanin* was reconstituted in
Escherichia coli. Cyanobacterial genes encoding enzymes required for the
conversion of heme to 3Z-phycocyanobilin, a precursor of phycobiliviolin
(namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin
oxidoreductase), were expressed from a plasmid under the control of the
hybrid trp-lac (trc) promoter. Genes for the apo-
phycoerythrocyanin.alpha. subunit (pecA) and the heterodimeric
lyase/isomerase (pecE and pecF), which catalyzes both the covalent
attachment of phycocyanobilin and its concurrent isomerization to
phycobiliviolin, were expressed from the trc promoter on a second plasmid.
Upon induction, recombinant E. coli used endogenous heme to produce
holo-PecA with absorbance and fluorescence properties similar to those of
the same protein produced in cyanobacteria. About two-thirds of the
apo-PecA was converted to holo-PecA. No significant bilin addition took
place in a similarly engineered E. coli strain that lacks pecE and pecF.
By using immobilized metal affinity chromatography, both apo-PecA and
holo-PecA were isolated as ternary complexes with PecE and PecF. The
identities of all three components in the ternary complexes were
established unambiguously by protein and tryptic peptide analyses
performed by matrix-assisted laser desorption ionization-time of flight
mass spectrometry.

ACCESSION NUMBER: 2002295599 EMBASE

TITLE: Biosynthesis of the cyanobacterial light-harvesting
polypeptide *phycoerythrocyanin* holo-.alpha.
subunit in a heterologous host.

AUTHOR: Tooley A.J.; Glazer A.N.

CORPORATE SOURCE: A.N. Glazer, Natural Reserve System, University of
California, 1111 Franklin Street, Oakland, CA 94607-5200,
United States. glazer@uclink4.berkeley.edu

SOURCE: Journal of Bacteriology, (2002) 184/17 (4666-4671).
Refs: 22

ISSN: 0021-9193 CODEN: JOBAAY

COUNTRY: United States

DOCUMENT TYPE: Journal; Article

FILE SEGMENT: 004 Microbiology

LANGUAGE: English

SUMMARY LANGUAGE: English

L6 ANSWER 31 OF 39 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.
TI Chromophore attachment to biliproteins: Specificity of PecE/PecF, a lyase-isomerase for the photoactive 3(1)-Cys-.alpha.84-phycoviolobilin chromophore of **phycoerythrocyanin**.
AB PecE and PecF, the products of two **phycoerythrocyanin** lyase genes (pecE and pecF) of Mastigocladus laminosus (*Fischerella*), catalyze two reactions: (1) the regiospecific addition of phycocyanobilin (PCB) to Cys-.alpha.84 of the **phycoerythrocyanin** .alpha.-subunit (PecA), and (2) the .DELTA.4.fwdarw..DELTA.2 isomerization of the PCB to the phycoviolobilin (PVB)-chromophore [Zhao et al. (2000) FEBS Lett. 469, 9-13]. The .alpha.-apoprotein (PecA) as well PecE and PecF were overexpressed from two strains of *M. laminosus*, with and without His-tags. The products of the spontaneous addition of PCB to PecA, and that of the reaction catalyzed by PecE/F, were characterized by their photochemistry and by absorption, fluorescence, circular dichroism of the four states obtained by irradiation with light (15-Z/E isomers of the chromophore) and/or modification of Cys-.alpha.98/99 with thiol-directed reagents. The spontaneous addition leads to a 3(1)-Cys-PCB adduct, which is characteristic of allophycocyanins and phycocyanins, while the addition catalyzed by PecE and PecF leads to a 3(1)-Cys-PVB adduct which after purification was identical to .alpha.-PEC. The specificity and kinetics of the chromophore additions were investigated with respect to the structure of the bilin substrate: The 3-ethylidene-bilins, viz., PCB, its 18-vinyl analogue phytochromobilin, phycoerythrobilin and its dimethylester, react spontaneously to yield the conventional addition products (3-H, 3(1)-Cys), while the 3-vinyl-substituted bilins, viz., bilirubin and biliverdin, were inactive. Only phycocyanobilin and phytochromobilin are substrates to the addition-isomerization reaction catalyzed by PecE/F. The slow spontaneous addition of phycoerythrobilin is not influenced, and there is in particular no catalyzed isomerization to urobilin.

ACCESSION NUMBER: 2001363892 EMBASE

TITLE: Chromophore attachment to biliproteins: Specificity of PecE/PecF, a lyase-isomerase for the photoactive 3(1)-Cys-.alpha.84-phycoviolobilin chromophore of **phycoerythrocyanin**.

AUTHOR: Storf M.; Parbel A.; Meyer M.; Strohmann B.; Scheer H.; Deng M.-G.; Zheng M.; Zhou M.; Zhao K.-H.

CORPORATE SOURCE: H. Scheer, Botanisches Institut, Universitat Munchen, Menzinger Strasse 67, D-80638 Munchen, Germany.

SOURCE: Biochemistry, (16 Oct 2001) 40/41 (12444-12456).
Refs: 88

ISSN: 0006-2960 CODEN: BICHAW

COUNTRY: United States

DOCUMENT TYPE: Journal; Article

FILE SEGMENT: 029 Clinical Biochemistry

LANGUAGE: English

SUMMARY LANGUAGE: English

L6 ANSWER 32 OF 39 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.

TI Novel activity of a **phycobiliprotein** lyase: Both the attachment of phycocyanobilin and the isomerization to phycoviolobilin are catalyzed by the proteins PecE and PecF encoded by the **phycoerythrocyanin** operon.

AB The structure of phycoviolobilin, the photoactive chromophore of .alpha.-**phycoerythrocyanin**, is incompatible with a chromophore ligation to the apoprotein via SH-addition (cysteine) to a .DELTA.3,31-double bond of the phycobilin. The two putative **phycoerythrocyanin** lyase genes of Mastigocladus laminosus, pecE and pecF, were overexpressed in *Escherichia coli*. Their action has been studied on the addition reaction of phycocyanobilin to apo-.alpha.-**phycoerythrocyanin** (PecA). In the absence of the components of .alpha.-PEC-phycoviolobilin lyase PecE

and PecF, or in the presence of only one of them, phycocyanobilin binds covalently to PecA forming a fluorescent chromoprotein with a red-shifted absorption ($\lambda_{max}=641$ nm) and low photoactivity (<10%). In the presence of both PecE and PecF, a chromoprotein forms which by its absorption ($\lambda_{max}=565$ nm) and high photoreversible photochromism (100% type I) has been identified as integral α -**phycoerythrocyanin**. We conclude that PecE and PecF jointly catalyze not only the addition of phycocyanobilin to PecA, but also its isomerization to the native phycoviolobilin chromophore. Copyright (C) 2000 Federation of European Biochemical Societies.

ACCESSION NUMBER: 2000081098 EMBASE

TITLE: Novel activity of a **phycobiliprotein** lyase: Both the attachment of phycocyanobilin and the isomerization to phycoviolobilin are catalyzed by the proteins PecE and PecF encoded by the **phycoerythrocyanin** operon.

AUTHOR: Zhao K.-H.; Deng M.-G.; Zheng M.; Zhou M.; Parbel A.; Storf M.; Meyer M.; Strohmann B.; Scheer H.

CORPORATE SOURCE: K.-H. Zhao, College of Life Sciences, Wuhan University, Wuhan 430072, China. scheer-h@botanik.biologie.uni-muenchen.de

SOURCE: FEBS Letters, (3 Mar 2000) 469/1 (9-13).

Refs: 41

ISSN: 0014-5793 CODEN: FEBBL

PUBLISHER IDENT.: S 0014-5793(00)01245-X

COUNTRY: Netherlands

DOCUMENT TYPE: Journal; Article

FILE SEGMENT: 004 Microbiology

LANGUAGE: English

SUMMARY LANGUAGE: English

L6 ANSWER 33 OF 39 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.

TI One century of protein crystallography: The phycobiliproteins.

AB The physical principles that underlay the rapid and efficient energy transfer from the light absorbing phycobilisomes to the reaction centre are conceivable from the knowledge of the exact three-dimensional structure of the phycobiliproteins and chromophores that are involved. The structure of the components and their assembly in the phycobilisomes could be determined by the structure analysis of X-ray data derived from **phycobiliprotein** crystals. Reports about these very aesthetic and brilliantly colored crystals have been published for more than a hundred years but it was only in the last decade that the structures of the different members of the **phycobiliprotein** family were solved for the first time at atomic resolution - all of them in Martinsried at the Max-Planck-Institut fur Biochemie. Despite the appearance of common structural principles the most important finding was that very subtle modifications in the structure and environment of the chromophores are sufficient to establish a highly specific light harvesting system in which the phycobiliproteins function with great cooperativity and efficiency.

ACCESSION NUMBER: 97164179 EMBASE

DOCUMENT NUMBER: 1997164179

TITLE: One century of protein crystallography: The phycobiliproteins.

AUTHOR: Betz M.

CORPORATE SOURCE: M. Betz, Max-Planck-Institut fur Biochemie, Abteilung Strukturforschung, Am Klopferspitz 18a, D-82152 Martinsried, Germany

SOURCE: Biological Chemistry, (1997) 378/3-4 (167-176).

Refs: 65

ISSN: 1431-6730 CODEN: BICHF3

COUNTRY: Germany

DOCUMENT TYPE: Journal; General Review

FILE SEGMENT: 004 Microbiology
029 Clinical Biochemistry

LANGUAGE: English

SUMMARY LANGUAGE: English

L6 ANSWER 34 OF 39 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.
TI A role for cpeYZ in cyanobacterial phycoerythrin biosynthesis.
AB Pigment mutant strain FdR1 of the filamentous cyanobacterium *Fremyella diplosiphon* is characterized by constitutive synthesis of the **phycobiliprotein** phycoerythrin due to insertional inactivation of the rcaC regulatory gene by endogenous transposon Tn5469. Whereas the parental strain Fd33 harbors five genomic copies of Tn5469, cells of strain FdR1 harbor six genomic copies of the element; the sixth copy in FdR1 is localized to the rcaC gene. Electroporation of FdR1 cells yielded secondary pigment mutant strains FdRIE1 and FdRIE4, which identically exhibited the FdR1 phenotype with significantly reduced levels of phycoerythrin. In both FdRIE1 and FdRIE4, a seventh genomic copy of Tn5469 was localized to the cpeY gene of the sequenced but phenotypically uncharacterized cpeYZ gene set. This gene set is located downstream of the cpeBA operon which encodes the α and β . subunits of phycoerythrin. Complementation experiments correlated cpeYZ activity to the phenotype of strains FdRIE1 and FdRIE4. The predicted CpeY and CpeZ proteins share significant sequence identity with the products of homologous cpeY and cpeZ genes reported for *Pseudanabaena* sp. strain PCC 7409 and *Synechococcus* sp. strain WH 8020, both of which synthesize phycoerythrin. The CpeY and CpeZ proteins belong to a family of structurally related cyanobacterial proteins that includes the subunits of the CpcE/CpcF phycocyanin α -subunit lyase of *Synechococcus* sp. strain PCC 7002 and the subunits of the PecE/PecF **phycoerythrocyanin** α -subunit lyase of *Anabaena* sp. strain PCC 7120. Phycobilisomes isolated from mutant strains FdRIE1 and FdRIE4 contained equal amounts of chromophorylated α . and β . subunits of phycoerythrin at 46% of the levels of the parental strain FdR1. These results suggest that the cpeYZ gene products function in phycoerythrin synthesis, possibly as a lyase involved in the attachment of phycoerythrobilin to the α . or β . subunit.

ACCESSION NUMBER: 97052785 EMBASE

DOCUMENT NUMBER: 1997052785

TITLE: A role for cpeYZ in cyanobacterial phycoerythrin biosynthesis.

AUTHOR: Kahn K.; Mazel D.; Houmard J.; De Marsac N.T.; Schaefer M.R.

CORPORATE SOURCE: M.R. Schaefer, School of Biological Sciences, University of Missouri, 5100 Rockhill Rd., Kansas City, MO 64110, United States. mschaefer@cctr.umkc.edu

SOURCE: Journal of Bacteriology, (1997) 179/4 (998-1006).
Refs: 36

ISSN: 0021-9193 CODEN: JOBAAY

COUNTRY: United States

DOCUMENT TYPE: Journal; Article

FILE SEGMENT: 004 Microbiology

LANGUAGE: English

SUMMARY LANGUAGE: English

L6 ANSWER 35 OF 39 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.

TI Candidate genes for the **phycoerythrocyanin** α . subunit lyase. Biochemical analysis of pecE and pecF interposon mutants.

AB The rod substructures of the *Anabaena* sp. PCC 7120 phycobilisome contain the light harvesting proteins C-phycocyanin and **phycoerythrocyanin** (PEC). Even at low light intensities, PEC represents no more than 5% of the phycobilisome protein. The β . subunits of both proteins carry thioether-linked phycocyanobilin (PCB) at β .-Cys-82 and β .-Cys-155; however, C-phycocyanin has PCB at α .-Cys-84 whereas PEC α . subunit carries phycobiliviolin at this position. The *Anabaena* sp. PCC 7120 pec operon is made up of five genes. PecB and pecA encode the β . and α . subunits of PEC, pecC encodes a linker polypeptide associated with PEC in the rod substructure, and pecE and pecF are genes of unknown function that show a high degree of homology to cpcE and cpcF,

that encode a C-phycocyanin .alpha. subunit PCB lyase (Fairchild, C. D., Zhao, J., Zhou, J., Colson, S. E., Bryant, D. A., and Glazer, A. N. (1992) Proc. Natl. Acad. Sci. U. S. A. 89, 7017-7021). Insertional mutants in pecE and pecF, and an interposon mutant in which a portion of both pecE and pecF was deleted, were constructed. All three types of mutants grew 1.3 times slower than wild-type under limiting light conditions and showed a 20% reduction in the PCB content of whole cells relative to chlorophyll .alpha.. Holo-PEC was missing from the phycobilisomes of all three types of mutants and the level of the PEC linker polypeptide was reduced relative to the wild-type. However, .apprx.30% of the wild-type level of the PEC .beta. subunit was present in all of these phycobilisomes. In contrast, the PEC .alpha. subunit was barely detectable in the pecE and pecF mutants, but was present in the pec EF deletion mutant as a PCB-adduct in a 1:1 ratio with the PEC .beta. subunit. The identity of this 'unnatural' adduct was confirmed by isolation of the subunit and amino-terminal sequencing. These biochemical results support the inference that pecE and pecF encode a PEC .alpha. subunit phycobiliviolin lyase, and, in conjunction with earlier findings, demonstrate that **phycobiliprotein** bilin lyases show high selectivity (rather than absolute specificity) for both the bilin and the polypeptide substrate.

ACCESSION NUMBER: 95162378 EMBASE

DOCUMENT NUMBER: 1995162378

TITLE: Candidate genes for the **phycoerythrocyanin** .alpha. subunit lyase. Biochemical analysis of pecE and pecF interposon mutants.

AUTHOR: Jung L.J.; Chan C.F.; Glazer A.N.

CORPORATE SOURCE: Stanley/Donner ASU, 229 Stanley Hall 3206, University of California, Berkeley, CA 94720-3206, United States

SOURCE: Journal of Biological Chemistry, (1995) 270/21
(12877-12884).

ISSN: 0021-9258 CODEN: JBCHA3

COUNTRY: United States

DOCUMENT TYPE: Journal; Article

FILE SEGMENT: 029 Clinical Biochemistry

LANGUAGE: English

SUMMARY LANGUAGE: English

L6 ANSWER 36 OF 39 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.

TI The complete amino acid sequence of R-phycocyanin-I .alpha. and .beta. subunits from the red alga Porphyridium cruentum. Structural and phylogenetic relationships of the phycocyanins within the **phycobiliprotein** families.

AB We present here the complete primary structure of R-phycocyanin-I .alpha. and .beta. subunits from the red alga Porphyridium cruentum. The .alpha. chain is composed of 162 amino acid residues (18049 Da, calculated from sequence, including chromophore) and carries a phycocyanobilin pigment covalently linked to Cys84. The .beta. chain contains 172 amino acids (19344 Da, calculated from sequence, including chromophores) and carries a phycocyanobilin pigment covalently linked at Cys82 and a phycoerythrobilin pigment at Cys153. A .gamma.-N-methyl asparagine residue was also characterised at position .beta.72 similar to other

phycobiliprotein .beta. subunits. R-phycocyanin-I from Porphyridium cruentum shares high sequence identity with C-phycocyanins (69-83%), R-phycocyanins (66-70%) and in a less extent with phycoerythrocyanins (57-65%) from various sources. The presented phylogenetic trees are based on a comparison of all **phycobiliprotein** amino acid sequences known so far and confirm the clear affiliation of the R-phycocyanins in the phycocyanin family. In spite of their particular phycobilin pattern, they do not represent intermediate forms between the phycocyanin and the phycoerythrin family.

Phycoerythrocyanin, a phycocyanin-related **phycobiliprotein** adapted to green light harvesting, is also shown to belong to the phycocyanin family. However, the phycoerythrocyanins diverge from phycocyanins in their different function and it is suggested that they

should be assigned to a separate group within the phycocyanin family.

ACCESSION NUMBER: 94116795 EMBASE
DOCUMENT NUMBER: 1994116795
TITLE: The complete amino acid sequence of R-phycocyanin-I .alpha. and .beta. subunits from the red alga Porphyridium cruentum. Structural and phylogenetic relationships of the phycocyanins within the **phycobiliprotein** families.
AUTHOR: Ducret A.; Sidler W.; Frank G.; Zuber H.
CORPORATE SOURCE: Inst. for Molec. Biology/Biophysics, Federal institute of Technology, CH-8093 Zurich, Switzerland
SOURCE: European Journal of Biochemistry, (1994) 221/1 (563-580).
ISSN: 0014-2956 CODEN: EJBCAI
COUNTRY: Germany
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English

L6 ANSWER 37 OF 39 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.
TI Structure of the genes encoding the rod-core linker polypeptides of Mastigocladus laminosus phycobilisomes and functional aspects of the **phycobiliprotein**/linker-polypeptide interactions.
AB The 3' portion of the cpc operon in Mastigocladus laminosus encloses the genes 5'-cpcF-cpcG1-cpcG2-cpcG3 3'. The three cpcG genes encode different phycocyanin-associated rod-core linker polypeptides of the phycobilisomes with predicted 279, 247 and 254 amino acids in length. The gene products CpcG show a high similarity at their N-terminal domains (190 amino acids) and an overall identity of 47-53% to one another. Each of the three CpcG polypeptides is highly related to one of the four CpcG gene products of Anabaena sp. PCC 7120 (66-81% identity). It is suggested that these pairs of rod-core linker polypeptides mediate the same specific type of phycocyanin .fwdarw. allophycocyanin interaction in the similar phycobilisomes of M. laminosus and Anabaena sp. PCC 7120. The similarity of the CpcG1, CpcG2 and CpcG3 polypeptides to the single CpcG rod-core linker polypeptide of Synechococcus sp. PCC 7002 (36-41% identity) is lower. The rod-core linker polypeptides are more distantly related to the rod linker polypeptides associated with phycocyanin or phycoerythrin. However, six conserved domains were identified within the N-terminal 190 amino acids of these linker proteins, which bear similar amino acid sequences, including highly conserved basic amino acids. A similar amino acid sequence but with conserved acidic amino acids can be found in the .beta. subunits of phycocyanin, phycoerythrin and **phycoerythrocyanin**, which is protruding into the central cavity of the **phycobiliprotein** hexamers. It is suggested that these domains are sites of **phycobiliprotein**-hexamer/rod and rod-core linker interactions.

ACCESSION NUMBER: 92141649 EMBASE
DOCUMENT NUMBER: 1992141649
TITLE: Structure of the genes encoding the rod-core linker polypeptides of Mastigocladus laminosus phycobilisomes and functional aspects of the **phycobiliprotein**/linker-polypeptide interactions.
AUTHOR: Glauser M.; Stirewalt V.L.; Bryant D.A.; Sidler W.; Zuber H.
CORPORATE SOURCE: Inst. fur Molekularbiol./Biophysik, ETH-Honggerberg-HPM, CH-8093 Zurich, Switzerland
SOURCE: European Journal of Biochemistry, (1992) 205/3 (927-937).
ISSN: 0014-2956 CODEN: EJBCAI
COUNTRY: Germany
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English

L6 ANSWER 38 OF 39 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.
TI Refined three-dimensional structure of **phycoerythrocyanin** from
the cyanobacterium *Mastigocladus laminosus* at 2.7 .ANG..
AB The structure of the **phycobiliprotein phycoerythrocyanin**
from the thermophilic cyanobacterium *Mastigocladus laminosus* has been
determined at 2.7 .ANG. resolution by X-ray diffraction methods on the
basis of the molecular model of C-phycocyanin from the same organism.
Hexagonal **phycoerythrocyanin** crystals of space group P63 with
cell constants a=b=156.86 .ANG., c=40.39 .ANG., alpha.=beta.=90.degree.,
gamma.=120.degree. are almost isomorphous to C-phycocyanin crystals. The
crystal structure has been refined by energy-restrained crystallographic
refinement and model building. The conventional crystallographic R-factor
of the final model was 19.2% with data to 2.7 .ANG. resolution. In
phycoerythrocyanin, the three (.alpha..beta.)-subunits are
arranged around a 3-fold symmetry axis, as in C-phycocyanin. The two
structures are very similar. After superposition, the 162 C(.alpha.) atoms
of the .alpha.-subunit have a mean difference of 0.71 .ANG. and the 171
C(.alpha.) atoms of the .beta.-subunit differ by 0.51 .ANG.. The
stereochemistry of the chiral atoms in the phycobiliviolin chromophore A84
is C((31))-R, C((4))-S. The configuration of the chromophore is C((10))-Z,
C((15))-Z and the conformation C((5))-anti, C((9))-syn and C((14))-anti
like the phycocyanobilin chromophores in **phycoerythrocyanin** and
C-phycocyanin.

ACCESSION NUMBER: 90086271 EMBASE
DOCUMENT NUMBER: 1990086271
TITLE: Refined three-dimensional structure of
phycoerythrocyanin from the cyanobacterium
Mastigocladus laminosus at 2.7 .ANG..
AUTHOR: Duerring M.; Huber R.; Bode W.; Ruembeli R.; Zuber H.
CORPORATE SOURCE: Max-Planck Institut fur Biochemie, D-8033 Martinsried,
Germany
SOURCE: Journal of Molecular Biology, (1990) 211/3 (633-644).
ISSN: 0022-2836 CODEN: JMOBAK
COUNTRY: United Kingdom
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English

L6 ANSWER 39 OF 39 WPIDS (C) 2003 THOMSON DERWENT
TI Analyzing soluble analyte concentration in sample, useful for diagnosing
disease, by performing competition assay using formed bodies to which are
attached at least one analyte, unlabeled and labeled ligands of analyte.
AN 2003-040599 [03] WPIDS
AB WO 200277645 A UPAB: 20030113
NOVELTY - Analyzing (M1) concentration of soluble analyte (I) comprising
utilizing formed bodies (II) to which analyte is bound, performing
competition assay in which unlabeled and labeled ligands (UL,LL) compete
for binding to (II)-bound analyte and (I), incubating control samples
containing (II) and varying known concentrations of LL with no UL,
analyzing test and control samples to obtain detectable signal from LL,
and using the data to determine concentration of (I), is new.

DETAILED DESCRIPTION - Analyzing (M1) the concentration or amount of
soluble analyte (I) in a sample, involves incubating test samples
containing an unknown concentration of (I), a predetermined amount of
formed bodies to which are attached at least one analyte, varying known
concentrations of unlabeled ligand (UL) that binds to the analyte, and a
known concentration of the ligand labeled with a detectable marker, where
LL and the UL compete for binding to the formed body-bound analyte and
(I); incubating several control samples which contains predetermined
amount of the formed bodies and varying known concentrations of LL with no
UL to permit binding to reach equilibrium, where the formed bodies are
between 1-100% coated with LL; analyzing test samples and control samples

in an instrument that measures detectable signal produced from LLs bound to the bound analytes on the formed bodies; identifying the intersection of a curve formed by plotting the signal against the concentration of LL in the control samples, and a second curve formed by plotting the signal against the concentration of total labeled and UL in the test samples; determining the concentration of UL that bound to (I) in the test samples by evaluating the difference D between the concentration that corresponds with the intersection and the constant LL concentration in the test samples; and determining the concentration of (I) in the sample by determining the product of the value D and the binding stoichiometry of the ligand to (I).

INDEPENDENT CLAIMS are also included for:

(1) a computer program that identifies and analyzes the amount of (I) in a sample by (M1);

(2) an analysis instrument that comprises an integrated computer program that identifies and analyzes the amount of (I) in a sample by implementing (M1); and

(3) a kit for performing a method of diagnosing a disease characterized by an altered level of (I) in the blood of a mammal, comprises components selected from ligands, detectable markers for labeling a ligand, formed bodies, suitable vessels for containing samples, suitable controls or tables of normal values of (I), instructions for performing the method and preparing the controls, suitable diluents and buffers for the samples, indicator charts for signal comparisons, disposable gloves, decontamination instructions, applicator sticks or containers, and/or sample preparator cups.

USE - (M1) is useful for analyzing concentration or amount of soluble analyte in a sample which is purified of any particles other than the analyte, and contains formed bodies having the analytes bound to it, and (I) shed from the formed bodies. The method is useful for analyzing

concentration or amount of soluble analyte which is a proteinaceous or chemical composition which can be naturally or covalently bound to a formed body, e.g. cell surface receptor, preferably an antigenic receptor anchored to a white blood cell by a glycosyl-phosphatidylinositol linkage.

(M1) is useful for analyzing the concentration or amount of soluble receptor from a sample which comprises formed bodies having the receptors bound to them, and the soluble receptors shed from the formed bodies, where the concentration of soluble receptors is calculated according to the formula D multiply binding valence of the ligand. (M1) is useful for diagnosing a disease characterized by an altered level of (I) in the blood of a mammal which involves analyzing the amount of (I) by performing (M1), comparing the concentration of (I) from the mammal's samples with known normal concentrations of (I) in samples of healthy mammalian blood, where a difference between the concentration of (I) in the mammal's blood sample and the normal concentration is indicative of disease. Preferably the method is useful for diagnosing a disease characterized by an altered level of soluble receptors shed by target cells having receptors bound to the cell surface into the blood of a mammal (all claimed). The method has medical applications such as diagnosis of diseases characterized by presence of certain analytes in a biological sample, or the evaluation of commercial or industrial samples containing analytes. The method is also

useful for monitoring the efficacy of treatment of various diseases characterized by shedding of bound receptors from target cells e.g. blood cells and other diseases in which the receptors or analytes are not naturally bound but are soluble. The method is useful for analyzing soluble and neutrophil-bound CD16b antigen, and for quantifying many other soluble and cell bound receptors, e.g. the CD100 receptor shed in the context of spinal cord injury. The method can be also be used to determine only (I), such as for thyroid factor T4 or soluble prostate surface antigen. The method is useful for distinguishing between the normal and diseased form of the prion protein, for quantifying non-biological analytes, e.g. for particular analytes in water or other fluids which the proteins are soluble.

ADVANTAGE - (M1) is simple, and enables rapid flow cytometric

analysis of samples. The method requires no pure or partially pure antigen, and lacks any requirement for an extraneous solid support for any component of the reaction. The elimination of these requirements allows the assays to be more easily performed in samples which contain preexisting formed bodies and receptors. The method is both simple and inexpensive requiring only a supply of purified unlabeled ligand and suitable fluorescent LL against the same analyte. The method does not involve lengthy incubation times or precautions regarding the use and disposal of radioactive material.

Dwg.1A/3

ACCESSION NUMBER: 2003-040599 [03] WPIDS
DOC. NO. NON-CPI: N2003-031888
DOC. NO. CPI: C2003-009585
TITLE: Analyzing soluble analyte concentration in sample, useful for diagnosing disease, by performing competition assay using formed bodies to which are attached at least one analyte, unlabeled and labeled ligands of analyte.
DERWENT CLASS: A89 B04 D16 S03
INVENTOR(S): SIIMAN, O
PATENT ASSIGNEE(S): (SIIM-I) SIIMAN O; (COUS) COULTER INT CORP
COUNTRY COUNT: 21
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 2002077645	A2	20021003	(200303)*	EN	45
RW: AT BE CH CY DE DK ES FI FR GB GR IE IT LU MC NL PT SE TR					
W: JP					
US 2002142289 A1 20021003 (200303)					

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 2002077645	A2	WO 2002-US3176	20020102
US 2002142289	A1	US 2001-768127	20010123

PRIORITY APPLN. INFO: US 2001-768127 20010123

=> s E. coli
L8 227457 E. COLI

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=> s 15 and 18
L9 9 L5 AND L8

=> d 19 ti abs ibib tot

L9 ANSWER 1 OF 9 MEDLINE
TI Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-alpha subunit in a heterologous host.
AB The entire pathway for the synthesis of a fluorescent holophycobiliprotein subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803) was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to the natural chromophore 3Z-phycocyanobilin, namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase, were expressed from a plasmid under control of the hybrid trp-lac (trc) promoter. Genes for the apoprotein (C-phycocyanin alpha subunit; cpcA) and the heterodimeric lyase (cpcE and cpcF) that catalyzes chromophore attachment were expressed from the trc promoter on a second plasmid. Upon induction, recombinant

E. coli used the cellular pool of heme to produce holo-CpcA with spectroscopic properties qualitatively and quantitatively similar to those of the same protein produced endogenously in cyanobacteria. About a third of the apo-CpcA was converted to holo-CpcA. No significant bilin addition took place in a similarly engineered **E. coli** strain that lacks cpcE and cpcF. This approach should permit incisive analysis of many remaining questions in **phycobiliprotein** biosynthesis. These studies also demonstrate the feasibility of generating constructs of these proteins *in situ* for use as fluorescent protein probes in living cells.

ACCESSION NUMBER: 2001504133 MEDLINE
DOCUMENT NUMBER: 21438034 PubMed ID: 11553806
TITLE: Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-alpha subunit in a heterologous host.
AUTHOR: Tooley A J; Cai Y A; Glazer A N
CORPORATE SOURCE: Department of Molecular and Cell Biology, University of California, 142 LSA no. 3200, Berkeley, CA 94720-3200, USA.
SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (2001 Sep 11) 98 (19) 10560-5.
Journal code: 7505876. ISSN: 0027-8424.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200111
ENTRY DATE: Entered STN: 20010913
Last Updated on STN: 20011105
Entered Medline: 20011101

L9 ANSWER 2 OF 9 MEDLINE
TI Expression of *Escherichia coli* phosphoenolpyruvate carboxylase in a cyanobacterium. Functional complementation of *Synechococcus* PCC 7942 ppc.
AB The gene (ppc) coding for phosphoenolpyruvate carboxylase (PEPCase) in the cyanobacterium *Synechococcus* PCC 7942 has been inactivated via insertional mutagenesis while being functionally complemented by *Escherichia coli* ppc. Cyanobacterial cells functionally complemented by *E. coli* ppc showed decreased PEPCase activity in crude cell lysates and detergent-permeabilized whole cell assays. Decreased rates of growth, reduced levels of chlorophyll a, and decreased photosynthetic O₂ evolution capacity per cell when compared to wild-type cyanobacterial cells were also observed. **Phycobiliprotein** levels were not affected. The results are discussed in terms of the impact of reduced PEPCase activity on cyanobacterial cell metabolism and the regulatory properties of the *E. coli* gene product.

ACCESSION NUMBER: 94105286 MEDLINE
DOCUMENT NUMBER: 94105286 PubMed ID: 8278492
TITLE: Expression of *Escherichia coli* phosphoenolpyruvate carboxylase in a cyanobacterium. Functional complementation of *Synechococcus* PCC 7942 ppc.
AUTHOR: Luinenburg I; Coleman J R
CORPORATE SOURCE: Department of Botany, University of Toronto, Ontario, Canada.
SOURCE: PLANT PHYSIOLOGY, (1993 Jan) 101 (1) 121-6.
Journal code: 0401224. ISSN: 0032-0889.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199402
ENTRY DATE: Entered STN: 19940218
Last Updated on STN: 19940218
Entered Medline: 19940208

L9 ANSWER 3 OF 9 MEDLINE

TI Isolation and characterization of the genes encoding allophycocyanin subunits and two linker proteins from *Synechocystis* 6714.
AB Genes encoding the phycobilisome core subunits allophycocyanin alpha and beta and a small core linker protein in *Synechocystis* sp. strain PCC 6714 were cloned and sequenced. These genes form an operon, apcABC, with a single transcription start site and two possible termination sites, one following apcB and the other following apcC. The promoter region, like those of the apcABC operons of other cyanobacteria, does not resemble the consensus promoter sequences of *Escherichia coli*. However, the apcABC promoters identified in four strains of cyanobacteria have conserved sequences centered at -50 and -10 with respect to the start of transcription. The apcE gene, encoding the protein that links the phycobilisome core to the thylakoid membrane, was also cloned from *Synechocystis* 6714 and sequenced. It is unlinked to the apcABC operon. As in other *Synechocystis* strains, the LCM polypeptide encoded by the apcE gene contains three repeats of the basic **phycobiliprotein** linker domain. The apcE gene promoter sequence bears little resemblance to either the *E. coli* consensus or the apcABC promoter region, but it is similar to the corresponding regions of other cyanobacterial apcE genes. In these cases, there are conserved sequences centered at -40 and -10 with respect to the transcription start site. These conserved promoter elements from the apcABC and apcE genes were also identified in the corresponding 5'-flanking regions of eleven transcript starts for cpc genes encoding phycocyanin subunits in cyanobacteria and algal chloroplasts. These results suggest that a factor yet to be described participates in transcription of **phycobiliprotein** genes.

ACCESSION NUMBER: 93222481 MEDLINE
DOCUMENT NUMBER: 93222481 PubMed ID: 8467079
TITLE: Isolation and characterization of the genes encoding allophycocyanin subunits and two linker proteins from *Synechocystis* 6714.
AUTHOR: DiMaggio L; Haselkorn R
CORPORATE SOURCE: Department of Chemistry and Molecular Genetics, University of Chicago, IL 60637.
CONTRACT NUMBER: GM 08282 (NIGMS)
SOURCE: PLANT MOLECULAR BIOLOGY, (1993 Mar) 21 (5) 835-45.
Journal code: 9106343. ISSN: 0167-4412.
PUB. COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-X06084; GENBANK-Z11906
ENTRY MONTH: 199305
ENTRY DATE: Entered STN: 19930521
Last Updated on STN: 19930521
Entered Medline: 19930511

L9 ANSWER 4 OF 9 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
TI Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-alpha subunit in a heterologous host.
AB The entire pathway for the synthesis of a fluorescent holophycobiliprotein subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803) was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to the natural chromophore 3Z-phycocyanobilin, namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase, were expressed from a plasmid under control of the hybrid trp-lac (trc) promoter. Genes for the apoprotein (C-phycocyanin alpha subunit; cpcA) and the heterodimeric lyase (cpcE and cpcF) that catalyzes chromophore attachment were expressed from the trc promoter on a second plasmid. Upon induction, recombinant *E. coli* used the cellular pool of heme to produce holo-CpcA with spectroscopic properties qualitatively and quantitatively similar to those of the same protein produced endogenously in cyanobacteria. About a third of the apo-CpcA was converted to holo-CpcA.

No significant bilin addition took place in a similarly engineered *E. coli* strain that lacks cpcE and cpcF. This approach should permit incisive analysis of many remaining questions in phycobiliprotein biosynthesis. These studies also demonstrate the feasibility of generating constructs of these proteins *in situ* for use as fluorescent protein probes in living cells.

ACCESSION NUMBER: 2001:482056 BIOSIS
DOCUMENT NUMBER: PREV200100482056
TITLE: Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-alpha subunit in a heterologous host.
AUTHOR(S): Tooley, Aaron J.; Cai, Yuping A.; Glazer, Alexander N. (1)
CORPORATE SOURCE: (1) Natural Reserve System, University of California System, 1111 Franklin Street, 6th Floor, Oakland, CA, 94607-5200: alexander.glazer@ucop.edu USA
SOURCE: Proceedings of the National Academy of Sciences of the United States of America, (September 11, 2001) Vol. 98, No. 19, pp. 10560-10565. print.
ISSN: 0027-8424.
DOCUMENT TYPE: Article
LANGUAGE: English
SUMMARY LANGUAGE: English

L9 ANSWER 5 OF 9 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
TI Isolation and characterization of the genes encoding allophycocyanin subunits and two linker proteins from *Synechocystis* 6714.
AB Genes encoding the phycobilisome core subunits allophycocyanin alpha and beta and a small core linker protein in *Synechocystis* sp. strain PCC 6714 were cloned and sequenced. These genes form an operon, apcABC, with a single transcription start site and two possible termination sites, one following apcB and the other following apcC. The promoter region, like those of the apcABC operons of other cyanobacteria, does not resemble the consensus promoter sequences of *Escherichia coli*. However, the apcABC promoters identified in four strains of cyanobacteria have conserved sequences centered at -50 and -10 with respect to the start of transcription. The apcE gene, encoding the protein that links the phycobilisome core to the thylakoid membrane, was also cloned from *Synechocystis* 6714 and sequenced. It is unlinked to the apcABC operon. As in other *Synechocystis* strains, the L-CM polypeptide encoded by the apcE gene contains three repeats of the basic phycobiliprotein linker domain. The apcE gene promoter sequence bears little resemblance to either the *E. coli* consensus or the apcABC promoter region, but it is similar to the corresponding regions of other cyanobacterial apcE genes. In these cases, there are conserved sequences centered at -40 and -10 with respect to the transcription start site. These conserved promoter elements from the apcABC and apcE genes were also identified in the corresponding 5'-flanking regions of eleven transcript starts for cpc genes encoding phycocyanin subunits in cyanobacteria and algal chloroplasts. These results suggest that a factor yet to be described participates in transcription of phycobiliprotein genes.

ACCESSION NUMBER: 1993:319341 BIOSIS
DOCUMENT NUMBER: PREV199396027691
TITLE: Isolation and characterization of the genes encoding allophycocyanin subunits and two linker proteins from *Synechocystis* 6714.
AUTHOR(S): Dimagno, Lisa; Haselkorn, Robert (1)
CORPORATE SOURCE: (1) Dep. Chem., Univ. Chicago, 920 East 58 St., Chicago, IL 60637 USA
SOURCE: Plant Molecular Biology, (1993) Vol. 21, No. 5, pp. 835-845.
ISSN: 0167-4412.
DOCUMENT TYPE: Article
LANGUAGE: English

L9 ANSWER 6 OF 9 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.

TI Expression of *Escherichia coli* phosphoenolpyruvate carboxylase in a cyanobacterium: Functional complementation of *Synechococcus* PCC 7942 ppc.
AB The gene (ppc) coding for phosphoenolpyruvate carboxylase (PEPCase) in the cyanobacterium *Synechococcus* PCC 7942 has been inactivated via insertional mutagenesis while being functionally complemented by *Escherichia coli* ppc. Cyanobacterial cells functionally complemented by *E. coli* ppc showed decreased PEPCase activity in crude cell lysates and detergent-permeabilized whole cell assays. Decreased rates of growth, reduced levels of chlorophyll a, and decreased photosynthetic O₂ evolution capacity per cell when compared to wild-type cyanobacterial cells were also observed. **Phycobiliprotein** levels were not affected. The results are discussed in terms of the impact of reduced PEPCase activity on cyanobacterial cell metabolism and the regulatory properties of the *E. coli* gene product.

ACCESSION NUMBER: 1993:164233 BIOSIS

DOCUMENT NUMBER: PREV199395085283

TITLE: Expression of *Escherichia coli* phosphoenolpyruvate carboxylase in a cyanobacterium: Functional complementation of *Synechococcus* PCC 7942 ppc.

AUTHOR(S): Luinenburg, Irene; Coleman, John R. (1)

CORPORATE SOURCE: (1) Dep. Bot., Univ. Toronto, Toronto, ON, Can. M5S 3B2

SOURCE: Plant Physiology (Rockville), (1993) Vol. 101, No. 1, pp. 121-126.

ISSN: 0032-0889.

DOCUMENT TYPE: Article

LANGUAGE: English

L9 ANSWER 7 OF 9 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.

TI Biosynthesis of the cyanobacterial light-harvesting polypeptide phycoerythrocyanin holo-.alpha. subunit in a heterologous host.

AB The entire pathway for the biosynthesis of the phycobiliviolin-bearing His-tagged holo-.alpha. subunit of the cyanobacterial photosynthetic accessory protein phycoerythrocyanin was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to 3Z-phycocyanobilin, a precursor of phycobiliviolin (namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase), were expressed from a plasmid under the control of the hybrid trp-lac (trc) promoter. Genes for the apo-phycoerythrocyanin .alpha. subunit (pecA) and the heterodimeric lyase/isomerase (pecE and pecF), which catalyzes both the covalent attachment of phycocyanobilin and its concurrent isomerization to phycobiliviolin, were expressed from the trc promoter on a second plasmid. Upon induction, recombinant *E. coli* used endogenous heme to produce holo-PecA with absorbance and fluorescence properties similar to those of the same protein produced in cyanobacteria. About two-thirds of the apo-PecA was converted to holo-PecA. No significant bilin addition took place in a similarly engineered *E. coli* strain that lacks pecE and pecF. By using immobilized metal affinity chromatography, both apo-PecA and holo-PecA were isolated as ternary complexes with PecE and PecF. The identities of all three components in the ternary complexes were established unambiguously by protein and tryptic peptide analyses performed by matrix-assisted laser desorption ionization-time of flight mass spectrometry.

ACCESSION NUMBER: 2002295599 EMBASE

TITLE: Biosynthesis of the cyanobacterial light-harvesting polypeptide phycoerythrocyanin holo-.alpha. subunit in a heterologous host.

AUTHOR: Tooley A.J.; Glazer A.N.

CORPORATE SOURCE: A.N. Glazer, Natural Reserve System, University of California, 1111 Franklin Street, Oakland, CA 94607-5200, United States. glazer@uclink4.berkeley.edu

SOURCE: Journal of Bacteriology, (2002) 184/17 (4666-4671).

Refs: 22

ISSN: 0021-9193 CODEN: JOBAAY

COUNTRY: United States

DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English

L9 ANSWER 8 OF 9 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.
TI Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-.alpha. subunit in a heterologous host.
AB The entire pathway for the synthesis of a fluorescent holophycobiliprotein subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803) was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to the natural chromophore 3Z-phycocyanobilin, namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase, were expressed from a plasmid under control of the hybrid trp-lac (trc) promoter. Genes for the apoprotein (C-phycocyanin .alpha. subunit; cpcA) and the heterodimeric lyase (cpcE and cpcF) that catalyzes chromophore attachment were expressed from the trc promoter on a second plasmid. Upon induction, recombinant *E. coli* used the cellular pool of heme to produce holo-CpcA with spectroscopic properties qualitatively and quantitatively similar to those of the same protein produced endogenously in cyanobacteria. About a third of the apo-CpcA was converted to holo-CpcA. No significant bilin addition took place in a similarly engineered *E. coli* strain that lacks cpcE and cpcF. This approach should permit incisive analysis of many remaining questions in phycobiliprotein biosynthesis. These studies also demonstrate the feasibility of generating constructs of these proteins *in situ* for use as fluorescent protein probes in living cells.

ACCESSION NUMBER: 2001329835 EMBASE
TITLE: Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-.alpha. subunit in a heterologous host.
AUTHOR: Tooley A.J.; Cai Y.A.; Glazer A.N.
CORPORATE SOURCE: A.N. Glazer, Natural Reserve System, University of California System, 1111 Franklin Street, Oakland, CA 94607-5200, United States. alexander.glazer@ucop.edu
SOURCE: Proceedings of the National Academy of Sciences of the United States of America, (11 Sep 2001) 98/19 (10560-10565).
Refs: 30
ISSN: 0027-8424 CODEN: PNASA6
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English

L9 ANSWER 9 OF 9 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.
TI Expression of phycobiliprotein genes in *Escherichia coli*.
AB The cpcA and cpcB genes, which encode the .alpha.- and .beta.-subunit apoproteins of *Synechococcus* 7002 phycocyanin (PC), respectively, are efficiently expressed in *Escherichia coli*. Studies performed with a series of deletions produced from the parental plasmid pAQPR1 suggest that transcription of these genes in *E. coli* is initiated from a promoter located not more than 384 bp upstream from the translation start of cpcB. An examination of the nucleotide sequence of this region reveals a consensus *E. coli* promoter centered approx. 374 bp upstream from the cpcB translation start. The cyanelle-encoded allophycocyanin .alpha.- and .beta.-subunit apoprotein genes (apcA and apcB) of *Cyanophora paradoxa* are also expressed in *E. coli*.

ACCESSION NUMBER: 86005229 EMBASE
DOCUMENT NUMBER: 1986005229
TITLE: Expression of phycobiliprotein genes in *Escherichia coli*.

AUTHOR: Bryant D.A.; Dubbs J.M.; Fields P.I.; et al.
CORPORATE SOURCE: Department of Molecular and Cell Biology, The Pennsylvania
State University, University Park, PA 16802, United States
SOURCE: FEMS Microbiology Letters, (1985) 29/3 (343-349).
CODEN: FMLED7
COUNTRY: Netherlands
DOCUMENT TYPE: Journal
FILE SEGMENT: 004 Microbiology
022 Human Genetics
LANGUAGE: English

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now available on STN
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NEWS 23 Sep 03 JAPIO has been reloaded and enhanced
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NEWS 25 Sep 16 CA Section Thesaurus available in CAPLUS and CA
NEWS 26 Oct 01 CASREACT Enriched with Reactions from 1907 to 1985
NEWS 27 Oct 21 EVENTLINE has been reloaded
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NEWS 30 Oct 25 MEDLINE SDI run of October 8, 2002
NEWS 31 Nov 18 DKILIT has been renamed APOLLIT
NEWS 32 Nov 25 More calculated properties added to REGISTRY
NEWS 33 Dec 02 TIBKAT will be removed from STN
NEWS 34 Dec 04 CSA files on STN
NEWS 35 Dec 17 PCTFULL now covers WP/PCT Applications from 1978 to date
NEWS 36 Dec 17 TOXCENTER enhanced with additional content
NEWS 37 Dec 17 Adis Clinical Trials Insight now available on STN
NEWS 38 Dec 30 ISMEC no longer available
NEWS 39 Jan 21 NUTRACEUT offering one free connect hour in February 2003
NEWS 40 Jan 21 PHARMAML offering one free connect hour in February 2003
NEWS 41 Jan 29 Simultaneous left and right truncation added to COMPENDEX,
ENERGY, INSPEC
NEWS 42 Feb 13 CANCERLIT is no longer being updated
NEWS 43 Feb 24 METADEX enhancements
NEWS 44 Feb 24 PCTGEN now available on STN
NEWS 45 Feb 24 TEMA now available on STN

NEWS 46 Feb 26 NTIS now allows simultaneous left and right truncation
NEWS 47 Feb 26 PCTFULL now contains images
NEWS 48 Mar 04 SDI PACKAGE for monthly delivery of multifile SDI results
NEWS 49 Mar 19 APOLLIT offering free connect time in April 2003
NEWS 50 Mar 20 EVENTLINE will be removed from STN
NEWS 51 Mar 24 PATDPAFULL now available on STN
NEWS 52 Mar 24 Additional information for trade-named substances without structures available in REGISTRY
NEWS 53 Mar 24 Indexing from 1957 to 1966 added to records in CA/CAPLUS

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=> s apo-enzyme

L1 565 APO-ENZYME

=> s holo-enzyme

L2 455 HOLO-ENZYME

=> s l2 and l1

L3 81 L2 AND L1

=> s phycobiliprotein

L4 1404 PHYCOBILIPROTEIN

=> s l4 and l2

L5 0 L4 AND L2

=> s l4 and holo

L6 11 L4 AND HOLO

=> d 16 ti abs ibib tot

L6 ANSWER 1 OF 11 MEDLINE

TI Biosynthesis of a fluorescent cyanobacterial C-phycocyanin **holo**-alpha subunit in a heterologous host.

AB The entire pathway for the synthesis of a fluorescent holophycobiliprotein subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803) was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to the natural chromophore 3Z-phycocyanobilin, namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase, were expressed from a plasmid under control of the hybrid trp-lac (trc) promoter. Genes for the apoprotein (C-phycocyanin alpha subunit; cpcA) and the heterodimeric lyase (cpcE and cpcF) that catalyzes chromophore attachment were expressed from the trc promoter on a second plasmid. Upon induction, recombinant *E. coli* used the cellular pool of heme to produce **holo**-CpcA with spectroscopic properties qualitatively and quantitatively similar to those of the same protein produced endogenously in cyanobacteria. About a third of the apo-CpcA was converted to **holo**-CpcA. No significant bilin addition took place in a similarly engineered *E. coli* strain that lacks cpcE and cpcF. This approach should permit incisive analysis of many remaining questions in **phycobiliprotein** biosynthesis. These studies also demonstrate the feasibility of generating constructs of these proteins *in situ* for use as fluorescent protein probes in living cells.

ACCESSION NUMBER: 2001504133 MEDLINE

DOCUMENT NUMBER: 21438034 PubMed ID: 11553806

TITLE: Biosynthesis of a fluorescent cyanobacterial C-phycocyanin **holo**-alpha subunit in a heterologous host.

AUTHOR: Tooley A J; Cai Y A; Glazer A N

CORPORATE SOURCE: Department of Molecular and Cell Biology, University of California, 142 LSA no. 3200, Berkeley, CA 94720-3200, USA:

SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (2001 Sep 11) 98 (19) 10560-5.
Journal code: 7505876. ISSN: 0027-8424.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

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ENTRY MONTH: 200111
ENTRY DATE: Entered STN: 20010913
Last Updated on STN: 20011105
Entered Medline: 20011101

L6 ANSWER 2 OF 11 MEDLINE
TI Candidate genes for the phycoerythrocyanin alpha subunit lyase.
Biochemical analysis of pecE and pecF interposon mutants.
AB The rod substructures of the Anabaena sp. PCC 7120 phycobilisome contain the light harvesting proteins C-phycocyanin and phycoerythrocyanin (PEC). Even at low light intensities, PEC represents no more than 5% of the phycobilisome protein. The beta subunits of both proteins carry thioether-linked phycocyanobilin (PCB) at beta-Cys-82 and beta-Cys-155; however, C-phycocyanin has PCB at alpha-Cys-84 whereas PEC alpha subunit carries phycobiliviolin at this position. The Anabaena sp. PCC 7120 pec operon is made up of five genes. PecB and pecA encode the beta and alpha subunits of PEC, pecC encodes a linker polypeptide associated with PEC in the rod substructure, and pecE and pecF are genes of unknown function that show a high degree of homology to cpcE and cpcF, that encode a C-phycocyanin alpha subunit PCB lyase (Fairchild, C. D., Zhao, J., Zhou, J., Colson, S. E., Bryant, D. A., and Glazer, A. N. (1992) Proc. Natl. Acad. Sci. U.S.A. 89, 7017-7021). Insertional mutants in pecE and pecF, and an interposon mutant in which a portion of both pecE and pecF was deleted, were constructed. All three types of mutants grew 1.3 times slower than wild-type under limiting light conditions and showed a 20% reduction in the PCB content of whole cells relative to chlorophyll alpha.
Holo-PEC was missing from the phycobilisomes of all three types of mutants and the level of the PEC linker polypeptide was reduced relative to the wild-type. However, approximately 30% of the wild-type level of the PEC beta subunit was present in all of these phycobilisomes. In contrast, the PEC alpha subunit was barely detectable in the pecE and pecF mutants, but was present in the pecEF deletion mutant as a PCB-adduct in a 1:1 ratio with the PEC beta subunit. The identity of this "unnatural" adduct was confirmed by isolation of the subunit and amino-terminal sequencing. These biochemical results support the inference that pecE and pecF encode a PEC alpha subunit phycobiliviolin lyase, and, in conjunction with earlier findings, demonstrate that **phycobiliprotein** bilin lyases show high selectivity (rather than absolute specificity) for both the bilin and the polypeptide substrate.

ACCESSION NUMBER: 95279433 MEDLINE
DOCUMENT NUMBER: 95279433 PubMed ID: 7759546
TITLE: Candidate genes for the phycoerythrocyanin alpha subunit lyase. Biochemical analysis of pecE and pecF interposon mutants.
AUTHOR: Jung L J; Chan C F; Glazer A N
CORPORATE SOURCE: Department of Molecular and Cell Biology, University of California, Berkeley 94720, USA.
CONTRACT NUMBER: GM28994 (NIGMS)
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (1995 May 26) 270 (21) 12877-84.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199506
ENTRY DATE: Entered STN: 19950707
Last Updated on STN: 19950707
Entered Medline: 19950628

L6 ANSWER 3 OF 11 USPATFULL
TI Engineering of living cells for the expression of holo-
phycobiliprotein-based constructs
AB Recombinant cells which express a fluorescent holo-

phycobiliprotein fusion protein and methods of use are described. The cells comprises a bilin, a recombinant bilin reductase, an apo-**phycobiliprotein** fusion protein precursor of the fusion protein comprising a corresponding apo-**phycobiliprotein** domain, and a recombinant **phycobiliprotein** domain-bilin lyase, which components react to form the **holo-phycobiliprotein** fusion protein. Also described are **holo-phycobiliprotein** based transcription reporter cells and assays, which cells conditionally express a heterologous-to-the-cell, fluorescent, first **holo-phycobiliprotein** domain.

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

ACCESSION NUMBER: 2003:37640 USPATFULL

TITLE: Engineering of living cells for the expression of **holo-phycobiliprotein**-based constructs

INVENTOR(S): Glazer, Alexander N., Berkeley, CA, UNITED STATES
Tooley, Aaron J., Berkeley, CA, UNITED STATES
Cai, Yuping, Carmel, IN, UNITED STATES

NUMBER	KIND	DATE
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PATENT INFORMATION: US 2003027285 A1 20030206

APPLICATION INFO.: US 2001-919486 A1 20010731 (9)

DOCUMENT TYPE: Utility

FILE SEGMENT: APPLICATION

LEGAL REPRESENTATIVE: RICHARD ARON OSMAN, SCIENCE AND TECHNOLOGY LAW GROUP,
75 DENISE DRIVE, HILLSBOROUGH, CA, 94010

NUMBER OF CLAIMS: 24

EXEMPLARY CLAIM: 1

NUMBER OF DRAWINGS: 3 Drawing Page(s)

LINE COUNT: 918

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

L6 ANSWER 4 OF 11 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.

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AB The entire pathway for the biosynthesis of the phycobiliviolin-bearing His-tagged **holo-.alpha.** subunit of the cyanobacterial photosynthetic accessory protein phycoerythrocyanin was reconstituted in Escherichia coli. Cyanobacterial genes encoding enzymes required for the conversion of heme to 3Z-phycocyanobilin, a precursor of phycobiliviolin (namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase), were expressed from a plasmid under the control of the hybrid trp-lac (trc) promoter. Genes for the apo-phycoerythrocyanin .alpha. subunit (pecA) and the heterodimeric lyase/isomerase (pecE and pecF), which catalyzes both the covalent attachment of phycocyanobilin and its concurrent isomerization to phycobiliviolin, were expressed from the trc promoter on a second plasmid. Upon induction, recombinant E. coli used endogenous heme to produce **holo-PecA** with absorbance and fluorescence properties similar to those of the same protein produced in cyanobacteria. About two-thirds of the apo-PecA was converted to **holo-PecA**. No significant bilin addition took place in a similarly engineered E. coli strain that lacks pecE and pecF. By using immobilized metal affinity chromatography, both apo-PecA and **holo-PecA** were isolated as ternary complexes with PecE and PecF. The identities of all three components in the ternary complexes were established unambiguously by protein and tryptic peptide analyses performed by matrix-assisted laser desorption ionization-time of flight mass spectrometry.

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CORPORATE SOURCE: A.N. Glazer, Natural Reserve System, University of California, 1111 Franklin Street, Oakland, CA 94607-5200, United States. glazer@uclink4.berkeley.edu
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Refs: 22
ISSN: 0021-9193 CODEN: JOBAAY

COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
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L6 ANSWER 5 OF 11 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.
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AUTHOR: Tooley A.J.; Cai Y.A.; Glazer A.N.

CORPORATE SOURCE: A.N. Glazer, Natural Reserve System, University of California System, 1111 Franklin Street, Oakland, CA 94607-5200, United States. alexander.glazer@ucop.edu
SOURCE: Proceedings of the National Academy of Sciences of the United States of America, (11 Sep 2001) 98/19 (10560-10565).

Refs: 30
ISSN: 0027-8424 CODEN: PNASA6

COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English

L6 ANSWER 6 OF 11 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.
TI Candidate genes for the phycoerythrocyanin .alpha. subunit lyase. Biochemical analysis of pecE and pecF interposon mutants.
AB The rod substructures of the *Anabaena* sp. PCC 7120 phycobilisome contain the light harvesting proteins C-phycocyanin and phycoerythrocyanin (PEC). Even at low light intensities, PEC represents no more than 5% of the phycobilisome protein. The .beta. subunits of both proteins carry thioether-linked phycocyanobilin (PCB) at .beta.-Cys-82 and .beta.-Cys-155; however, C-phycocyanin has PCB at .alpha.-Cys-84 whereas PEC .alpha. subunit carries phycobiliviolin at this position. The *Anabaena* sp. PCC 7120 pec operon is made up of five genes. PecB and pecA encode the .beta. and .alpha. subunits of PEC, pecC encodes a linker polypeptide

associated with PEC in the rod substructure, and pecE and pecF are genes of unknown function that show a high degree of homology to cpcE and cpcF, that encode a C-phycocyanin .alpha. subunit PCB lyase (Fairchild, C. D., Zhao, J., Zhou, J., Colson, S. E., Bryant, D. A., and Glazer, A. N. (1992) Proc. Natl. Acad. Sci. U. S. A. 89, 7017-7021). Insertional mutants in pecE and pecF, and an interposon mutant in which a portion of both pecE and pecF was deleted, were constructed. All three types of mutants grew 1.3 times slower than wild-type under limiting light conditions and showed a 20% reduction in the PCB content of whole cells relative to chlorophyll .alpha.. Holo-PEC was missing from the phycobilisomes of all three types of mutants and the level of the PEC linker polypeptide was reduced relative to the wild-type. However, apprx.30% of the wild-type level of the PEC .beta. subunit was present in all of these phycobilisomes. In contrast, the PEC .alpha. subunit was barely detectable in the pecE and pecF mutants, but was present in the pec EF deletion mutant as a PCB-adduct in a 1:1 ratio with the PEC .beta. subunit. The identity of this 'unnatural' adduct was confirmed by isolation of the subunit and amino-terminal sequencing. These biochemical results support the inference that pecE and pecF encode a PEC .alpha. subunit phycobiliviolin lyase, and, in conjunction with earlier findings, demonstrate that **phycobiliprotein** bilin lyases show high selectivity (rather than absolute specificity) for both the bilin and the polypeptide substrate.

ACCESSION NUMBER: 95162378 EMBASE

DOCUMENT NUMBER: 1995162378

TITLE: Candidate genes for the phycoerythrocyanin .alpha. subunit lyase. Biochemical analysis of pecE and pecF interposon mutants.

AUTHOR: Jung L.J.; Chan C.F.; Glazer A.N.

CORPORATE SOURCE: Stanley/Donner ASU, 229 Stanley Hall 3206, University of California,Berkeley, CA 94720-3206, United States

SOURCE: Journal of Biological Chemistry, (1995) 270/21 (12877-12884).

ISSN: 0021-9258 CODEN: JBCHA3

COUNTRY: United States

DOCUMENT TYPE: Journal; Article

FILE SEGMENT: 029 Clinical Biochemistry

LANGUAGE: English

SUMMARY LANGUAGE: English

L6 ANSWER 7 OF 11 HCPLUS COPYRIGHT 2003 ACS

TI Engineering of living cells for the expression of **holo-phycobiliprotein-based constructs**

AB Recombinant cells which express a fluorescent **holo-phycobiliprotein** fusion protein and methods of use are described. The cells comprises a bilin, a recombinant bilin reductase, an apo-**phycobiliprotein** fusion protein precursor of the fusion protein comprising a corresponding apo-**phycobiliprotein** domain, and a recombinant **phycobiliprotein** domain-bilin lyase, which components react to form the **holo-phycobiliprotein** fusion protein. Also described are **holo-phycobiliprotein** based transcription reporter cells and assays, which cells conditionally express a heterologous-to-the-cell, fluorescent, first **holo-phycobiliprotein** domain.

ACCESSION NUMBER: 2003:97917 HCPLUS

DOCUMENT NUMBER: 138:148684

TITLE: Engineering of living cells for the expression of **holo-phycobiliprotein-based constructs**

INVENTOR(S): Glazer, Alexander N.; Tooley, Aaron J.; Cai, Yuping

PATENT ASSIGNEE(S): USA

SOURCE: U.S. Pat. Appl. Publ., 13 pp.

CODEN: USXXCO

DOCUMENT TYPE: Patent

LANGUAGE: English

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 2003027285	A1	20030206	US 2001-919486	20010731
WO 2003012448	A1	20030213	WO 2002-US24245	20020730

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

PRIORITY APPLN. INFO.: US 2001-919486 A 20010731

L6 ANSWER 8 OF 11 HCAPLUS COPYRIGHT 2003 ACS

TI Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-.alpha. subunit in a heterologous host

AB The entire pathway for the synthesis of a fluorescent holophycobiliprotein subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803) was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to the natural chromophore 3Z-phycocyanobilin, namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase, were expressed from a plasmid under control of the hybrid trp-lac (trc) promoter. Genes for the apoprotein (C-phycocyanin .alpha. subunit; cpcA) and the heterodimeric lyase (cpcE and cpcF) that catalyzes chromophore attachment were expressed from the trc promoter on a second plasmid. Upon induction, recombinant *E. coli* used the cellular pool of heme to produce holo-CpcA with spectroscopic properties qual. and quant. similar to those of the same protein produced endogenously in cyanobacteria. About a third of the apo-CpcA was converted to holo-CpcA. No significant bilin addn. took place in a similarly engineered *E. coli* strain that lacks cpcE and cpcF. This approach should permit incisive anal. of many remaining questions in phycobiliprotein biosynthesis. These studies also demonstrate the feasibility of generating constructs of these proteins in situ for use as fluorescent protein probes in living cells.

ACCESSION NUMBER: 2001:705481 HCAPLUS

DOCUMENT NUMBER: 136:2664

TITLE: Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-.alpha. subunit in a heterologous host

AUTHOR(S): Tooley, Aaron J.; Cai, Yuping A.; Glazer, Alexander N.

CORPORATE SOURCE: Department of Molecular and Cell Biology, University of California, Berkeley, CA, 94720-3200, USA

SOURCE: Proceedings of the National Academy of Sciences of the United States of America (2001), 98(19), 10560-10565

CODEN: PNASA6; ISSN: 0027-8424

PUBLISHER: National Academy of Sciences

DOCUMENT TYPE: Journal

LANGUAGE: English

REFERENCE COUNT: 30 THERE ARE 30 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L6 ANSWER 9 OF 11 HCAPLUS COPYRIGHT 2003 ACS

TI Candidate genes for the phycoerythrocyanin .alpha. subunit lyase and biochemical analysis of pecE and pecF interposon mutants

AB The rod substructures of the *Anabaena* sp. PCC 7120 phycobilisome contain the light harvesting proteins C-phycocyanin and phycoerythrocyanin (PEC).

Even at low light intensities, PEC represents no more than 5% of the phycobilisome protein. The .beta. subunits of both proteins carry thioether-linked phycocyanobilin (PCB) at .beta.-Cys-82 and .beta.-Cys-155; however, C-phycocyanin has PCB at .alpha.-Cys-84 whereas PEC .alpha. subunit carries phycobiliviolin at this position. The *Anabaena* sp. PCC 7120 pec operon is made up of five genes. PecB and pecA encode the .beta. and .alpha. subunits of PEC, pecC encodes a linker polypeptide assocd. with PEC in the rod substructure, and pecE and pecF are genes of unknown function that show a high degree of homol. to cpcE and cpcF, that encode a C-phycocyanin .alpha. subunit PCB lyase (Fairchild, C. D., Zhao, J., Zhou, J., Colson, S. E., Bryant, D. A., and Glazer, A. N. (1992) Proc. Natl. Acad. Sci. U.S.A. 89, 7017-7021). Insertional mutants in pecE and pecF, and an interposon mutant in which a portion of both pecE and pecF was deleted, were constructed. All three types of mutants grew 1.3 times slower than wild-type under limiting light conditions and showed a 20% redn. in the PCB content of whole cells relative to chlorophyll a. Holo-PEC was missing from the phycobilisomes of all three types of mutants and the level of the PEC linker polypeptide was reduced relative to the PEC linker polypeptide was reduced relative to the wild-type. However, .apprx.30% of the wild-type level of the PEC .beta. subunit was present in all of these phycobilisomes. In contrast, the PEC .alpha. subunit was barely detectable in the pecE and pecF mutants, but was present in the pecEF deletion mutant as a PCB-adduct in a 1:1 ratio with the PEC .beta. subunit. The identity of this "unnatural" adduct was confirmed by isolation of the subunit and amino-terminal sequencing. These biochem. results support the inference that pecE and pecF encode a PEC .alpha. subunit phycobiliviolin lyase, and, in conjunction with earlier findings, demonstrate that phycobiliprotein bilin lyases show high selectivity (rather than abs. specificity) for both the bilin and the polypeptide substrate.

ACCESSION NUMBER: 1995:597221 HCAPLUS
DOCUMENT NUMBER: 123:250826
TITLE: Candidate genes for the phycoerythrocyanin .alpha. subunit lyase and biochemical analysis of pecE and pecF interposon mutants
AUTHOR(S): Jung, Linda J.; Chan, Crystal F.; Glazer, Alexander N.
CORPORATE SOURCE: Department Molecular Cell Biology, University California, Berkeley, CA, 94720, USA
SOURCE: Journal of Biological Chemistry (1995), 270(21), 12877-84
CODEN: JBCHA3; ISSN: 0021-9258
PUBLISHER: American Society for Biochemistry and Molecular Biology
DOCUMENT TYPE: Journal
LANGUAGE: English

L6 ANSWER 10 OF 11 SCISEARCH COPYRIGHT 2003 ISI (R)
TI Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo -alpha subunit in a heterologous host
AB The entire pathway for the synthesis of a fluorescent

holophycobiliprotein subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803) was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to the natural chromophore 3Z-phycocyanobilin, namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase, were expressed from a plasmid under control of the hybrid trp-lac (trc) promoter. Genes for the apoprotein (C-phycocyanin alpha subunit; cpcA) and the heterodimeric lyase (cpcE and cpcF) that catalyzes chromophore attachment were expressed from the trc promoter on a second plasmid. Upon induction, recombinant *E. coli* used the cellular pool of heme to produce holo-CpcA with spectroscopic properties qualitatively and quantitatively similar to those of the same protein produced endogenously in cyanobacteria. About a third of the apo-CpcA was converted to holo-CpcA. No significant bilin

addition took place in a similarly engineered *E. coli* strain that lacks cpcE and cpcF. This approach should permit incisive analysis of many remaining questions in **phycobiliprotein** biosynthesis. These studies also demonstrate the feasibility of generating constructs of these proteins *in situ* for use as fluorescent protein probes in living cells.

ACCESSION NUMBER: 2001:754757 SCISEARCH

THE GENUINE ARTICLE: 472CZ

TITLE: Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-alpha subunit in a heterologous host

AUTHOR: Tooley A J; Cai Y P A; Glazer A N (Reprint)

CORPORATE SOURCE: Univ Calif Syst, Nat Res Syst, 1111 Franklin St, 6th Floor, Oakland, CA 94607 USA (Reprint); Univ Calif Berkeley, Dept Mol & Cell Biol, Berkeley, CA 94720 USA

COUNTRY OF AUTHOR: USA

SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (11 SEP 2001) Vol. 98, No. 19, pp. 10560-10565.

Publisher: NATL ACAD SCIENCES, 2101 CONSTITUTION AVE NW, WASHINGTON, DC 20418 USA.

ISSN: 0027-8424.

DOCUMENT TYPE: Article; Journal

LANGUAGE: English

REFERENCE COUNT: 28

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

L6 ANSWER 11 OF 11 SCISEARCH COPYRIGHT 2003 ISI (R)

TI CANDIDATE GENES FOR THE PHYCOERYTHROCYANIN ALPHA-SUBUNIT LYASE - BIOCHEMICAL-ANALYSIS OF PECE AND PECF INTERPOSON MUTANTS

AB The rod substructures of the *Anabaena* sp, PCC 7120 phycobilisome contain the light harvesting proteins C-phycocyanin and phycoerythrocyanin (PEC). Even at low light intensities, PEC represents no more than 5% of the phycobilisome protein. The beta subunits of both proteins carry thioether-linked phycocyanobilin (PCB) at beta-Cys-82 and beta-Cys-155; however, C-phycocyanin has PCB at alpha-Cys-84 whereas PEC alpha subunit carries phycobiliviolin at this position. The *Anabaena* sp, PCC 7120 pec operon is made up of five genes, PecB and pecA encode the beta and alpha subunits of PEC, pecC encodes a linker polypeptide associated with PEC in the rod substructure, and pecE and pecF are genes of unknown function that show a high degree of homology to cpcE and cpcF, that encode a C-phycocyanin alpha subunit PCB lyase (Fairchild, C., D., Zhao, J., Zhou, J., Colson, S., E., Bryant, D. A., and Glazer, A. N. (1992) Proc. Natl. Acad. Sci. U.S. A. 89, 7017-7021). Insertional mutants in pecE and pecF, and an interposon mutant in which a portion of both pecE and pecF was deleted, were constructed. All three types of mutants grew 1.3 times slower than wild-type under limiting light conditions and showed a 20% reduction in the PCB content of whole cells relative to chlorophyll alpha. Holo-PEC was missing from the phycobilisomes of all three types of mutants and the level of the PEC linker polypeptide was reduced relative to the wild-type. However, similar to 30% of the wild-type level of the PEC beta subunit was present in all of these phycobilisomes. In contrast, the PEC alpha subunit was barely detectable in the pecE and pecF mutants, but was present in the pecEF deletion mutant as a PCB-adduct in a 1:1 ratio with the PEC beta subunit. The identity of this "unnatural" adduct was confirmed by isolation of the subunit and amino-terminal sequencing. These biochemical results support the inference that pecE and pecF encode a PEC a subunit phycobiliviolin lyase, and, in conjunction with earlier findings, demonstrate that **phycobiliprotein** bilin lyases show high selectivity (rather than absolute specificity) for both the bilin and the polypeptide substrate.

ACCESSION NUMBER: 95:367157 SCISEARCH

THE GENUINE ARTICLE: QZ711

TITLE: CANDIDATE GENES FOR THE PHYCOERYTHROCYANIN ALPHA-SUBUNIT LYASE - BIOCHEMICAL-ANALYSIS OF PECE AND PECF INTERPOSON MUTANTS

AUTHOR: JUNG L J; CHAN C F; GLAZER A N (Reprint)
CORPORATE SOURCE: UNIV CALIF BERKELEY, DEPT MOLEC & CELL BIOL, 229 STANLEY
HALL, BERKELEY, CA, 94720 (Reprint); UNIV CALIF BERKELEY,
DEPT MOLEC & CELL BIOL, BERKELEY, CA, 94720
COUNTRY OF AUTHOR: USA
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (26 MAY 1995) Vol. 270,
No. 21, pp. 12877-12884.
ISSN: 0021-9258.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE
LANGUAGE: ENGLISH
REFERENCE COUNT: 39
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

=> d his

(FILE 'HOME' ENTERED AT 12:57:47 ON 28 MAR 2003)

FILE 'MEDLINE, USPATFULL, DGENE, EMBASE, WPIDS, BIOBUSINESS, HCAPLUS,
SCISEARCH, JICST-EPLUS, FSTA' ENTERED AT 12:59:31 ON 28 MAR 2003

L1 565 S APO-ENZYME
L2 455 S HOLO-ENZYME
L3 81 S L2 AND L1
L4 1404 S PHYCOBILIPROTEIN
L5 0 S L4 AND L2
L6 11 S L4 AND HOLO

=> s l4 and apo
L7 32 L4 AND APO

=> s l6 and l7
L8 7 L6 AND L7

=> d l8 ti abs ibib tot

L8 ANSWER 1 OF 7 MEDLINE
TI Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo
-alpha subunit in a heterologous host.
AB The entire pathway for the synthesis of a fluorescent holophycobiliprotein
subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803)
was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding
enzymes required for the conversion of heme to the natural chromophore
3Z-phycocyanobilin, namely, heme oxygenase 1 and 3Z-
phycocyanobilin:ferredoxin oxidoreductase, were expressed from a plasmid
under control of the hybrid trp-lac (trc) promoter. Genes for the
apoprotein (C-phycocyanin alpha subunit; cpcA) and the heterodimeric lyase
(cpcE and cpcF) that catalyzes chromophore attachment were expressed from
the trc promoter on a second plasmid. Upon induction, recombinant *E. coli*
used the cellular pool of heme to produce holo-CpcA with
spectroscopic properties qualitatively and quantitatively similar to those
of the same protein produced endogenously in cyanobacteria. About a third
of the apo-CpcA was converted to holo-CpcA. No
significant bilin addition took place in a similarly engineered *E. coli*
strain that lacks cpcE and cpcF. This approach should permit incisive
analysis of many remaining questions in phycobiliprotein
biosynthesis. These studies also demonstrate the feasibility of generating
constructs of these proteins *in situ* for use as fluorescent protein probes
in living cells.

ACCESSION NUMBER: 2001504133 MEDLINE
DOCUMENT NUMBER: 21438034 PubMed ID: 11553806
TITLE: Biosynthesis of a fluorescent cyanobacterial C-phycocyanin
holo-alpha subunit in a heterologous host.
AUTHOR: Tooley A J; Cai Y A; Glazer A N

CORPORATE SOURCE: Department of Molecular and Cell Biology, University of California, 142 LSA no. 3200, Berkeley, CA 94720-3200, USA.
SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (2001 Sep 11) 98 (19) 10560-5.
Journal code: 7505876. ISSN: 0027-8424.

PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200111
ENTRY DATE: Entered STN: 20010913
Last Updated on STN: 20011105
Entered Medline: 20011101

L8 ANSWER 2 OF 7 USPATFULL

TI Engineering of living cells for the expression of **holo-phycobiliprotein**-based constructs

AB Recombinant cells which express a fluorescent **holo-phycobiliprotein** fusion protein and methods of use are described. The cells comprises a bilin, a recombinant bilin reductase, an **apo-phycobiliprotein** fusion protein precursor of the fusion protein comprising a corresponding **apo-phycobiliprotein** domain, and a recombinant **phycobiliprotein** domain-bilin lyase, which components react to form the **holo-phycobiliprotein** fusion protein. Also described are **holo-phycobiliprotein** based transcription reporter cells and assays, which cells conditionally express a heterologous-to-the-cell, fluorescent, first **holo-phycobiliprotein** domain.

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

ACCESSION NUMBER: 2003:37640 USPATFULL
TITLE: Engineering of living cells for the expression of **holo-phycobiliprotein**-based constructs
INVENTOR(S): Glazer, Alexander N., Berkeley, CA, UNITED STATES
Tooley, Aaron J., Berkeley, CA, UNITED STATES
Cai, Yuping, Carmel, IN, UNITED STATES

	NUMBER	KIND	DATE
PATENT INFORMATION:	US 2003027285	A1	20030206
APPLICATION INFO.:	US 2001-919486	A1	20010731 (9)
DOCUMENT TYPE:	Utility		
FILE SEGMENT:	APPLICATION		
LEGAL REPRESENTATIVE:	RICHARD ARON OSMAN, SCIENCE AND TECHNOLOGY LAW GROUP, 75 DENISE DRIVE, HILLSBOROUGH, CA, 94010		
NUMBER OF CLAIMS:	24		
EXEMPLARY CLAIM:	1		
NUMBER OF DRAWINGS:	3 Drawing Page(s)		
LINE COUNT:	918		

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

L8 ANSWER 3 OF 7 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.
TI Biosynthesis of the cyanobacterial light-harvesting polypeptide phycoerythrocyanin **holo-.alpha.** subunit in a heterologous host.
AB The entire pathway for the biosynthesis of the phycobiliviolin-bearing His-tagged **holo-.alpha.** subunit of the cyanobacterial photosynthetic accessory protein phycoerythrocyanin was reconstituted in Escherichia coli. Cyanobacterial genes encoding enzymes required for the conversion of heme to 3Z-phycocyanobilin, a precursor of phycobiliviolin (namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase), were expressed from a plasmid under the control of the hybrid trp-lac (trc) promoter. Genes for the **apo**

-phycoerythrocyanin .alpha. subunit (pecA) and the heterodimeric lyase/isomerase (pecE and pecF), which catalyzes both the covalent attachment of phycocyanobilin and its concurrent isomerization to phycobiliviolin, were expressed from the trc promoter on a second plasmid. Upon induction, recombinant E. coli used endogenous heme to produce holo-PecA with absorbance and fluorescence properties similar to those of the same protein produced in cyanobacteria. About two-thirds of the apo-PecA was converted to holo-PecA. No significant bilin addition took place in a similarly engineered E. coli strain that lacks pecE and pecF. By using immobilized metal affinity chromatography, both apo-PecA and holo-PecA were isolated as ternary complexes with PecE and PecF. The identities of all three components in the ternary complexes were established unambiguously by protein and tryptic peptide analyses performed by matrix-assisted laser desorption ionization-time of flight mass spectrometry.

ACCESSION NUMBER: 2002295599 EMBASE
TITLE: Biosynthesis of the cyanobacterial light-harvesting polypeptide phycoerythrocyanin holo-.alpha. subunit in a heterologous host.
AUTHOR: Tooley A.J.; Glazer A.N.
CORPORATE SOURCE: A.N. Glazer, Natural Reserve System, University of California, 1111 Franklin Street, Oakland, CA 94607-5200, United States. glazer@uclink4.berkeley.edu
SOURCE: Journal of Bacteriology, (2002) 184/17 (4666-4671).
Refs: 22
ISSN: 0021-9193 CODEN: JOBAAY
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English

L8 ANSWER 4 OF 7 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.
TI Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-.alpha. subunit in a heterologous host.
AB The entire pathway for the synthesis of a fluorescent holophycobiliprotein subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803) was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to the natural chromophore 3Z-phycocyanobilin, namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase, were expressed from a plasmid under control of the hybrid trp-lac (trc) promoter. Genes for the apoprotein (C-phycocyanin .alpha. subunit; cpcA) and the heterodimeric lyase (cpcE and cpcF) that catalyzes chromophore attachment were expressed from the trc promoter on a second plasmid. Upon induction, recombinant E. coli used the cellular pool of heme to produce holo-CpcA with spectroscopic properties qualitatively and quantitatively similar to those of the same protein produced endogenously in cyanobacteria. About a third of the apo-CpcA was converted to holo-CpcA. No significant bilin addition took place in a similarly engineered E. coli strain that lacks cpcE and cpcF. This approach should permit incisive analysis of many remaining questions in phycobiliprotein biosynthesis. These studies also demonstrate the feasibility of generating constructs of these proteins *in situ* for use as fluorescent protein probes in living cells.

ACCESSION NUMBER: 2001329835 EMBASE
TITLE: Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-.alpha. subunit in a heterologous host.
AUTHOR: Tooley A.J.; Cai Y.A.; Glazer A.N.
CORPORATE SOURCE: A.N. Glazer, Natural Reserve System, University of California System, 1111 Franklin Street, Oakland, CA 94607-5200, United States. alexander.glazer@ucop.edu
SOURCE: Proceedings of the National Academy of Sciences of the United States of America, (11 Sep 2001) 98/19

(10560-10565).

Refs: 30

ISSN: 0027-8424 CODEN: PNASA6

COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English

L8 ANSWER 5 OF 7 HCAPLUS COPYRIGHT 2003 ACS
TI Engineering of living cells for the expression of holo-phycobiliprotein-based constructs
AB Recombinant cells which express a fluorescent holo-phycobiliprotein fusion protein and methods of use are described. The cells comprises a bilin, a recombinant bilin reductase, an apo-phycobiliprotein fusion protein precursor of the fusion protein comprising a corresponding apo-phycobiliprotein domain, and a recombinant phycobiliprotein domain-bilin lyase, which components react to form the holo-phycobiliprotein fusion protein. Also described are holo-phycobiliprotein based transcription reporter cells and assays, which cells conditionally express a heterologous-to-the-cell, fluorescent, first holo-phycobiliprotein domain.

ACCESSION NUMBER: 2003:97917 HCAPLUS
DOCUMENT NUMBER: 138:148684
TITLE: Engineering of living cells for the expression of holo-phycobiliprotein-based constructs
INVENTOR(S): Glazer, Alexander N.; Tooley, Aaron J.; Cai, Yuping
PATENT ASSIGNEE(S): USA
SOURCE: U.S. Pat. Appl. Publ., 13 pp.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 2003027285	A1	20030206	US 2001-919486	20010731
WO 2003012448	A1	20030213	WO 2002-US24245	20020730
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				

PRIORITY APPLN. INFO.: US 2001-919486 A 20010731

L8 ANSWER 6 OF 7 HCAPLUS COPYRIGHT 2003 ACS
TI Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-.alpha. subunit in a heterologous host
AB The entire pathway for the synthesis of a fluorescent holophycobiliprotein subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803) was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to the natural chromophore 3Z-phycocyanobilin, namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase, were expressed from a plasmid under control of the hybrid trp-lac (trc) promoter. Genes for the apoprotein (C-phycocyanin .alpha. subunit; cpcA) and the heterodimeric

lyase (*cpcE* and *cpcF*) that catalyzes chromophore attachment were expressed from the *trc* promoter on a second plasmid. Upon induction, recombinant *E. coli* used the cellular pool of heme to produce **holo-CpcA** with spectroscopic properties qual. and quant. similar to those of the same protein produced endogenously in cyanobacteria. About a third of the **apo-CpcA** was converted to **holo-CpcA**. No significant bilin addn. took place in a similarly engineered *E. coli* strain that lacks *cpcE* and *cpcF*. This approach should permit incisive anal. of many remaining questions in **phycobiliprotein** biosynthesis. These studies also demonstrate the feasibility of generating constructs of these proteins *in situ* for use as fluorescent protein probes in living cells.

ACCESSION NUMBER: 2001:705481 HCAPLUS
DOCUMENT NUMBER: 136:2664
TITLE: Biosynthesis of a fluorescent cyanobacterial C-phycocyanin **holo**-alpha. subunit in a heterologous host
AUTHOR(S): Tooley, Aaron J.; Cai, Yuping A.; Glazer, Alexander N.
CORPORATE SOURCE: Department of Molecular and Cell Biology, University of California, Berkeley, CA, 94720-3200, USA
SOURCE: Proceedings of the National Academy of Sciences of the United States of America (2001), 98(19), 10560-10565
PUBLISHER: National Academy of Sciences
DOCUMENT TYPE: Journal
LANGUAGE: English
REFERENCE COUNT: 30 THERE ARE 30 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L8 ANSWER 7 OF 7 SCISEARCH COPYRIGHT 2003 ISI (R)
TI Biosynthesis of a fluorescent cyanobacterial C-phycocyanin **holo**-alpha subunit in a heterologous host
AB The entire pathway for the synthesis of a fluorescent holophycobiliprotein subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803) was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to the natural chromophore 3Z-phycocyanobilin, namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase, were expressed from a plasmid under control of the hybrid *trp-lac* (*trc*) promoter. Genes for the apoprotein (C-phycocyanin alpha subunit; *cpcA*) and the heterodimeric lyase (*cpcE* and *cpcF*) that catalyzes chromophore attachment were expressed from the *trc* promoter on a second plasmid. Upon induction, recombinant *E. coli* used the cellular pool of heme to produce **holo-CpcA** with spectroscopic properties qualitatively and quantitatively similar to those of the same protein produced endogenously in cyanobacteria. About a third of the **apo-CpcA** was converted to **holo-CpcA**. No significant bilin addition took place in a similarly engineered *E. coli* strain that lacks *cpcE* and *cpcF*. This approach should permit incisive analysis of many remaining questions in **phycobiliprotein** biosynthesis. These studies also demonstrate the feasibility of generating constructs of these proteins *in situ* for use as fluorescent protein probes in living cells.

ACCESSION NUMBER: 2001:754757 SCISEARCH
THE GENUINE ARTICLE: 472CZ
TITLE: Biosynthesis of a fluorescent cyanobacterial C-phycocyanin **holo**-alpha subunit in a heterologous host
AUTHOR: Tooley A J; Cai Y P A; Glazer A N (Reprint)
CORPORATE SOURCE: Univ Calif Syst, Nat Res Syst, 1111 Franklin St, 6th Floor, Oakland, CA 94607 USA (Reprint); Univ Calif Berkeley, Dept Mol & Cell Biol, Berkeley, CA 94720 USA
COUNTRY OF AUTHOR: USA
SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (11 SEP 2001) Vol. 98, No. 19, pp. 10560-10565.
Publisher: NATL ACAD SCIENCES, 2101 CONSTITUTION AVE NW,

WASHINGTON, DC 20418 USA.

ISSN: 0027-8424.

DOCUMENT TYPE: Article; Journal

LANGUAGE: English

REFERENCE COUNT: 28

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

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(FILE 'HOME' ENTERED AT 12:57:47 ON 28 MAR 2003)

FILE 'MEDLINE, USPATFULL, DGENE, EMBASE, WPIDS, BIOBUSINESS, HCAPLUS,
SCISEARCH, JICST-EPLUS, FSTA' ENTERED AT 12:59:31 ON 28 MAR 2003

L1	565 S APO-ENZYME
L2	455 S HOLO-ENZYME
L3	81 S L2 AND L1
L4	1404 S PHYCOBILIPROTEIN
L5	0 S L4 AND L2
L6	11 S L4 AND HOLO
L7	32 S L4 AND APO
L8	7 S L6 AND L7

CC 7-2 (Enzymes)
 AB Unavailable
 ST phytochromobilin synthase oat; Avena phytochromobilin synthase
 IT Oat
 (purifn. and characterization of phytochromobilin synthase from Avena sativa)
 IT 138263-99-7P, Phytochromobilin synthase
 RL: BAC (Biological activity or effector, except adverse); BSU (Biological study, unclassified); PRP (Properties); PUR (Purification or recovery); BIOL (Biological study); PREP (Preparation)
 (purifn. and characterization of phytochromobilin synthase from Avena sativa)

L92 ANSWER 5 OF 5 HCAPLUS COPYRIGHT 2003 ACS
 AN 1992:36617 HCAPLUS
 DN 116:36617
 TI **Holophytochrome** assembly. Coupled assay for phytochromobilin synthase in organello
 AU Terry, Matthew J.; Lagarias, J. Clark
 CS Dep. Biochem. Biophys., Univ. California, Davis, CA, 95616, USA
 SO Journal of Biological Chemistry (1991), 266(33), 22215-21
 CODEN: JBCHA3; ISSN: 0021-9258
 DT Journal
 LA English
 CC 7-1 (Enzymes)
 Section cross-reference(s): 6, 11
 AB Utilizing an in vitro coupled assay system, the authors show that isolated plastids from cucumber cotyledons convert the linear tetrapyrrole biliverdin IX. α . to the free phytochrome chromophore, phytochromobilin, which assembles with oat apophytochrome to yield photoactive **holoprotein**. The spectral properties of this synthetic phytochrome are indistinguishable from those of the natural photoreceptor. The plastid-dependent biliverdin conversion activity is strongly stimulated by both NADPH and ATP. Substitution of the nonnatural XIII. α . isomer of biliverdin for the IX. α . isomer affords a synthetic **holophytochrome** adduct with blue-shifted difference spectra. These results, together with expts. using boiled plastids, indicate that phytochromobilin synthesis from biliverdin is enzyme-mediated. Expts. where NADPH (and ATP) levels in intact developing chloroplasts are manipulated by feeding the metabolites 3-phosphoglycerate, dihydroxyacetone phosphate, and glucose 6-phosphate or by illumination with white light, support the hypothesis that the enzyme that accomplishes this conversion, phytochromobilin synthase, is plastid-localized. It is therefore likely that all of the enzymes of the phytochrome chromophore biosynthetic pathway reside in the plastid.
 ST phytochrome assembly etioplast chloroplast plant; phytochromobilin synthase detection plastid plant
 IT Oat
 (apophytochrome of seedling of, phytochromobilin of cucumber cotyledon and synthetic biliverdin XIII. α .-contg. phytochromobilin binding by)

IT **Phytochromes**
 RL: SPN (Synthetic preparation); PREP (Preparation)
 (biliverdin XIII. α .-contg., prepn. and properties of)

IT Light
 (phytochrome formation by cucumber cotyledon chloroplast response to)

IT Chloroplast
 (phytochrome synthase detection in and phytochrome assembly by, of plant)

IT Cucumber
 (phytochromobilin of plastid of, formation of and oat apophytochrome binding by)

IT Plant

IT (phytochromobilin synthase in plastids of and phytochrome assembly by)
Phytochromes
 RL: FORM (Formation, nonpreparative)
 (Pr, formation of, by plant plastid)

IT **Phytochromes**
 RL: ANST (Analytical study)
 (apo-, phytochromobilin of cucumber cotyledon and synthetic biliverdin
 XIII.alpha.-contg. phytochromobilin coupling with, of oat seedling)

IT Plastid
 (etio-, phytochrome synthase detection in and phytochrome assembly by,
 of plant)

IT **138263-99-7**, Phytochromobilin synthase
 RL: ANT (Analyte); ANST (Analytical study)
 (detection of, in chloroplast and etioplast of plant, by coupled assay)

IT 78249-71-5, Phytochromobilin
 RL: ANST (Analytical study)
 (formation of and apophytochrome of plant coupling with, in plant
 chloroplast and etioplast)

IT 56-65-5, 5'-ATP, biological studies
 RL: BIOL (Biological study)
 (phytochromobilin synthase of plant chloroplast and etioplast
 stimulation by)

IT **114-25-0**, Biliverdin IX.alpha. 28022-06-2, Biliverdin
 XIII.alpha.
 RL: RCT (Reactant); RACT (Reactant or reagent)
 (reaction of, with phytochromobilin synthase of plant etioplast and
 chloroplast)

=> sel hit rn
 E9 THROUGH E15 ASSIGNED

=> fil reg
 FILE 'REGISTRY' ENTERED AT 13:56:10 ON 27 MAR 2003
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 PLEASE SEE "HELP USAGETERMS" FOR DETAILS.
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Property values tagged with IC are from the ZIC/VINITI data file
 provided by InfoChem.

STRUCTURE FILE UPDATES: 26 MAR 2003 HIGHEST RN 500755-46-4
 DICTIONARY FILE UPDATES: 26 MAR 2003 HIGHEST RN 500755-46-4

TSCA INFORMATION NOW CURRENT THROUGH MAY 20, 2002

Please note that search-term pricing does apply when
 conducting SmartSELECT searches.

Crossover limits have been increased. See HELP CROSSOVER for details.

Experimental and calculated property data are now available. See HELP PROPERTIES for more information. See STNote 27, Searching Properties in the CAS Registry File, for complete details:
<http://www.cas.org/ONLINE/STN/STNOTES/stnotes27.pdf>

=> s e9-e15 not 180,181
 1 138263-99-7/BI
 (138263-99-7/RN)
 1 114-25-0/BI
 (114-25-0/RN)
 1 18097-67-1/BI
 (18097-67-1/RN)

1 20298-86-6/BI
(20298-86-6/RN)
1 347401-12-1/BI
(347401-12-1/RN)
1 347401-20-1/BI
(347401-20-1/RN)
1 347401-21-2/BI
(347401-21-2/RN)
L93 3 (138263-99-7/BI OR 114-25-0/BI OR 18097-67-1/BI OR 20298-86-6/BI
OR 347401-12-1/BI OR 347401-20-1/BI OR 347401-21-2/BI) NOT
(L80 OR L81)

=> d ide can tot

L93 ANSWER 1 OF 3 REGISTRY COPYRIGHT 2003 ACS
RN 347401-20-1 REGISTRY
CN Oxidoreductase, ferredoxin:15,16-dihydrobiliverdin (9CI) (CA INDEX NAME)
OTHER NAMES:
CN Ferredoxin:15,16-dihydrobiliverdin oxidoreductase
MF Unspecified
CI MAN
SR CA
LC STN Files: CA, CAPLUS

*** STRUCTURE DIAGRAM IS NOT AVAILABLE ***
3 REFERENCES IN FILE CA (1962 TO DATE)
3 REFERENCES IN FILE CAPLUS (1962 TO DATE)

REFERENCE 1: 137:365572

REFERENCE 2: 136:50278

REFERENCE 3: 135:73274

L93 ANSWER 2 OF 3 REGISTRY COPYRIGHT 2003 ACS
RN 138263-99-7 REGISTRY
CN Synthase, phytochromobilin (9CI) (CA INDEX NAME)
OTHER NAMES:
CN Ferredoxin:3Z-phytochromobilin oxidoreductase
CN Phytochromobilin synthase
MF Unspecified
CI MAN
SR CA
LC STN Files: AGRICOLA, BIOSIS, CA, CAPLUS

*** STRUCTURE DIAGRAM IS NOT AVAILABLE ***
8 REFERENCES IN FILE CA (1962 TO DATE)
8 REFERENCES IN FILE CAPLUS (1962 TO DATE)

REFERENCE 1: 137:365572

REFERENCE 2: 136:50278

REFERENCE 3: 135:207297

REFERENCE 4: 135:118588

REFERENCE 5: 135:73274

REFERENCE 6: 132:204723

REFERENCE 7: 125:190274

REFERENCE 8: 116:36617

L93 ANSWER 3 OF 3 REGISTRY COPYRIGHT 2003 ACS

RN 114-25-0 REGISTRY

CN 21H-Biline-8,12-dipropanoic acid, 3,18-diethenyl-1,19,22,24-tetrahydro-2,7,13,17-tetramethyl-1,19-dioxo-3,18-divinyl- (9CI) (CA INDEX NAME)

OTHER CA INDEX NAMES:

CN Biline-8,12-dipropionic acid, 1,19,22,24-tetrahydro-2,7,13,17-tetramethyl-1,19-dioxo-3,18-divinyl- (8CI)

CN Pyrrole-3-propionic acid, 2-[3-(2-carboxyethyl)-4-methyl-5-[(3-methyl-5-oxo-4-vinyl-3-pyrrolin-2-ylidene)methyl]-2H-pyrrol-2-ylidene]methyl]-4-methyl-5-[(4-methyl-5-oxo-3-vinyl-3-pyrrolin-2-ylidene)methyl]- (7CI)

OTHER NAMES:

CN Biliverdin

CN Biliverdin IX. α .

CN Biliverdine

CN Dehydrobilirubin

CN Oocyan

CN Protopobiliverdin IX. α .

CN Uteroverdine

FS STEREOSEARCH

DR 493-89-0, 27818-05-9, 29575-14-2, 189246-93-3

MF C33 H34 N4 O6

CI COM

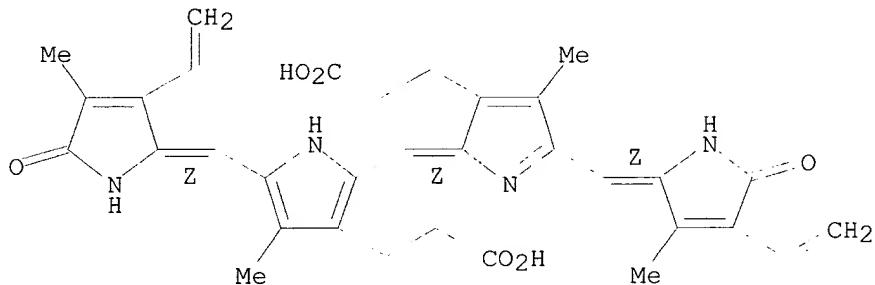
LC STN Files: AGRICOLA, ANABSTR, BEILSTEIN*, BIOBUSINESS, BIOSIS, BIOTECHNO, CA, CABA, CANCERLIT, CAOLD, CAPLUS, CASREACT, CHEMLIST, CSCHEM, DDFU, DRUGU, EMBASE, GMELIN*, HODOC*, IPA, MEDLINE, MRCK*, TOXCENTER, USPATFULL

(*File contains numerically searchable property data)

Other Sources: EINECS**, NDSL**, TSCA**

(**Enter CHEMLIST File for up-to-date regulatory information)

Double bond geometry as shown.



PROPERTY DATA AVAILABLE IN THE 'PROP' FORMAT

614 REFERENCES IN FILE CA (1962 TO DATE)

43 REFERENCES TO NON-SPECIFIC DERIVATIVES IN FILE CA

614 REFERENCES IN FILE CAPLUS (1962 TO DATE)

1 REFERENCES IN FILE CAOLD (PRIOR TO 1967)

REFERENCE 1: 138:182973

REFERENCE 2: 138:165503

REFERENCE 3: 138:148684

REFERENCE 4: 138:120036

REFERENCE 5: 138:39129

REFERENCE 6: 138:34738
 REFERENCE 7: 138:21090
 REFERENCE 8: 138:2706
 REFERENCE 9: 138:174
 REFERENCE 10: 137:365572

=> d his

(FILE 'HOME' ENTERED AT 12:54:49 ON 27 MAR 2003)
 SET COST OFF

FILE 'HCAPLUS' ENTERED AT 12:55:07 ON 27 MAR 2003
 E US20030027285/PN

L1 1 S E3
 E GLAZER A/AU
 L2 262 S E3,E7,E11,E13,E14
 E TOOLEY A/AU
 L3 3 S E4
 E CAI Y/AU
 L4 253 S E3-E18
 E CAI YU/AU
 L5 30 S E3,E10
 E CAI YUPING/AU
 L6 15 S E3
 L7 4 S E4
 E BILIPROTEIN/CT
 E E4+ALL
 L8 6658 S E7,E5+NT
 L9 2 S HOLOPHYCOCOBILIPROTEIN OR HOLO() (PHYCOBILIPROTEIN OR PHYCOBILI#

FILE 'REGISTRY' ENTERED AT 13:01:53 ON 27 MAR 2003

L10 1 S 9059-22-7
 L11 1 S 14875-96-8

FILE 'HCAPLUS' ENTERED AT 13:02:57 ON 27 MAR 2003

L12 9974 S L10 OR L11
 L13 2648 S HEME OXYGENASE
 L14 30884 S HEME OR PROTOHEME OR REDUCED HEMATIN OR HEM FE OR FERROHEME O
 L15 66 S L8 AND L12-L14
 L16 2 S L9 AND L15
 L17 813 S PHYCOCOBILIPROTEIN
 L18 6658 S L8,L9
 L19 66 S L18 AND L12-L14
 L20 6 S L19 AND (RECOMBIN? OR CONSTRUCT)
 L21 126 S L2-L7 AND L8,L9,L17,L18
 L22 4 S L12-L14 AND L21
 L23 302 S BILIN
 L24 336 S ?PHYCOCYANOBILIN?
 L25 816 S ?PHYCOCOBILIPROTEIN?
 L26 122 S ?PHYCOERYTHROCYANIN?
 L27 12 S ?PHYCOCOBILIVIOLIN?
 L28 173 S ?PHYCOERYTHROBILIN?
 L29 2074 S ?PHYCOERYTHRIN?
 L30 112 S L2-L7 AND L23-L29
 L31 4 S L12-L14 AND L30
 L32 7 S L1,L9,L16,L20,L22,L31
 L33 6 S L32 NOT PHARMACEUTICAL/TI

SEL RN L1

FILE 'REGISTRY' ENTERED AT 13:20:58 ON 27 MAR 2003

L34 9 S E1-E9
 L35 1 S 20298-86-6
 L36 1 S 93527-36-7
 L37 45 S C33H38N4O6/MF AND NC4/ES AND 4/NR
 L38 44 S L37 AND BILIN?
 L39 7 S L38 AND 8 12 DIPROPANOIC AND 18 ETHYL 3 ETHYLIDENE
 L40 7 S L39 AND 2 7 13 17 TETRAMETHYL 1 19 DIOXO
 L41 7 S L35, L36, L40
 L42 1 S 347401-12-1
 E PHYCOCYANOBILIN/CN
 L43 1 S E8
 E APO-PHYCOBILIPROTEIN/CN
 E PHYCOCYANIN/CN
 L44 116 S PHYCOCYANIN (L) ALPHA (L) SUBUNIT
 L45 1 S 168680-20-4
 L46 1 S 124861-40-1
 L47 1 S 18097-67-1
 L48 174 S C33H38N4O6/MF
 L49 45 S L48 AND NC4/ES AND 4/NR
 L50 3 S L49 AND BILINE 8 12 DIPROPANOIC AND 18 ETHENYL 3 ETHYLIDENE A
 L51 3 S L47, L50
 L52 1 S 347401-21-2

FILE 'HCAPLUS' ENTERED AT 13:37:26 ON 27 MAR 2003

L53 350 S L41 OR PHYCOCYANOBILIN
 L54 8 S L42 OR PHYCOCYANOBILIN(S) FERREDOXIN(S) OXIDOREDUCTASE
 L55 8 S L45 OR PHYCOERYTHROCYANIN LYASE
 L56 12 S L46 OR PHYCOBILIVIOLIN
 L57 25 S L12-L14 AND L53
 L58 43 S L54-L57
 L59 31 S L58 AND L8
 L60 2 S L59 AND L9

FILE 'REGISTRY' ENTERED AT 13:41:42 ON 27 MAR 2003

L61 1 S 144378-42-7

FILE 'HCAPLUS' ENTERED AT 13:42:39 ON 27 MAR 2003

L62 6 S L61 OR PHYCOCYANIN (S) ALPHA(S) SUBUNIT (S) PHYCOCYANOBILIN LY
 L63 28 S L54-L56, L62
 L64 7 S L63 AND L12-L14
 L65 76 S HOLO? AND L8, L53-L60, L62-L64
 L66 71 S L65 AND (PD<=20010731 OR PRD<=20010731 OR AD<=20010731)
 L67 7 S L54, L55, L62 AND L66
 L68 19 S L54, L55, L62
 L69 19 S L67, L68
 L70 36 S L2-L7 AND L53-L60, L62-L63
 L71 4 S L70 AND L66
 L72 5 S L70 AND L65
 L73 5 S L71, L72
 E GENETIC ENGINEERING/CT
 E E3+ALL
 L74 79989 S E2+NT
 L75 10 S L74 AND L65
 L76 12 S L73, L75
 L77 263 S L74 AND L12-L14
 L78 8 S L77 AND L23-L29, L53-L56, L62, L63
 L79 14 S L76, L78

FILE 'HCAPLUS' ENTERED AT 13:50:25 ON 27 MAR 2003

SEL HIT RN

FILE 'REGISTRY' ENTERED AT 13:50:40 ON 27 MAR 2003
L80 8 S E1-E8
L81 10 S L41,L42,L43,L45-L47,L51,L52,L61 NOT L80

FILE 'HCAPLUS' ENTERED AT 13:52:50 ON 27 MAR 2003
L82 7 S L42,L52
L83 1 S L82 NOT L79
L84 1 S L83 AND L1-L33,L53-L60,L62-L79,L82-L83

FILE 'REGISTRY' ENTERED AT 13:54:24 ON 27 MAR 2003
L85 2 S 138263-99-7 OR 347401-20-1
L86 2 S 114-25-0 OR 18097-67-1

FILE 'HCAPLUS' ENTERED AT 13:54:36 ON 27 MAR 2003
L87 8 S L85
L88 753 S L86
L89 5 S L87 AND L1-L33,L53-L60,L62-L69,L82-L84,L87,L88 NOT L79
L90 5 S L84,L89
L91 2 S L90 AND L88
L92 5 S L90,L91

FILE 'HCAPLUS' ENTERED AT 13:55:52 ON 27 MAR 2003
SEL HIT RN

FILE 'REGISTRY' ENTERED AT 13:56:10 ON 27 MAR 2003
L93 3 S E9-E15 NOT L80,L81

=> fil biosis
FILE 'BIOSIS' ENTERED AT 14:00:43 ON 27 MAR 2003
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FILE COVERS 1969 TO DATE.
CAS REGISTRY NUMBERS AND CHEMICAL NAMES (CNS) PRESENT
FROM JANUARY 1969 TO DATE.

RECORDS LAST ADDED: 26 March 2003 (20030326/ED)

=> d all tot 1105

L105 ANSWER 1 OF 4 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
AN 2002:464629 BIOSIS
DN PREV200200464629
TI Biosynthesis of the cyanobacterial light-harvesting polypeptide
phycoerythrocyanin holo-alpha subunit in a heterologous
host.
AU **Tooley, Aaron J.; Glazer, Alexander N. (1)**
CS (1) Natural Reserve System, University of California, 1111 Franklin
Street, 6th Floor, Oakland, CA, 94607-5200: glazer@uclink4.berkeley.edu
USA
SO Journal of Bacteriology, (September, 2002) Vol. 184, No. 17, pp.
4666-4671. <http://intl-jb.asm.org/>. print.
ISSN: 0021-9193.
DT Article
LA English
AB The entire pathway for the biosynthesis of the **phycobiliviolin**
-bearing His-tagged **holo-alpha** subunit of the cyanobacterial
photosynthetic accessory protein **phycoerythrocyanin** was
reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes
required for the conversion of heme to 3Z-**phycocyanobilin**, a
precursor of **phycobiliviolin** (namely, heme oxygenase 1 and 3Z-
phycocyanobilin:ferredoxin oxidoreductase), were expressed from a

plasmid under the control of the hybrid trp-lac (trc) promoter. Genes for the apo-**phycoerythrocyanin** alpha subunit (pecA) and the heterodimeric lyase/isomerase (pecE and pecF), which catalyzes both the covalent attachment of **phycocyanobilin** and its concurrent isomerization to **phycobiliviolin**, were expressed from the trc promoter on a second plasmid. Upon induction, recombinant E. coli used endogenous heme to produce **holo-PecA** with absorbance and fluorescence properties similar to those of the same protein produced in cyanobacteria. About two-thirds of the apo-PecA was converted to **holo-PecA**. No significant **bilin** addition took place in a similarly engineered E. coli strain that lacks pecE and pecF. By using immobilized metal affinity chromatography, both apo-PecA and **holo-PecA** were isolated as ternary complexes with PecE and PecF. The identities of all three components in the ternary complexes were established unambiguously by protein and tryptic peptide analyses performed by matrix-assisted laser desorption ionization-time of flight mass spectrometry.

CC Genetics and Cytogenetics - General *03502
 Physiology and Biochemistry of Bacteria *31000
 Genetics of Bacteria and Viruses *31500

BC Enterobacteriaceae 06702
 Nostocaceae 09241

IT Major Concepts
 Molecular Genetics (Biochemistry and Molecular Biophysics)

IT Chemicals & Biochemicals
 apo-PecA protein; **holo-PecA** protein;
 phycoerythrocyanin: biosynthesis, **holo-alpha** subunit,
 light-harvesting polypeptide

IT Methods & Equipment
 immobilized metal affinity chromatography: purification method;
 matrix-assisted laser desorption ionization-time of flight mass
 spectrometry: analytical method; tryptic peptide analysis: analytical
 method

ORGN Super Taxa
 Enterobacteriaceae: Facultatively Anaerobic Gram-Negative Rods,
 Eubacteria, Bacteria, Microorganisms; Nostocaceae: Nostocales,
 Cyanobacteria, Oxygenic Photosynthetic Bacteria, Eubacteria, Bacteria,
 Microorganisms

ORGN Organism Name
 Anabaena sp. (Nostocaceae): strain-PCC7120; Escherichia coli
 (Enterobacteriaceae): DH5-alpha

ORGN Organism Superterms
 Bacteria; Cyanobacteria; Eubacteria; Microorganisms

GEN Anabaena pecA gene [Anabaena apo-**phycoerythrocyanin**
 alpha-subunit gene] (Nostocaceae); Anabaena pecE gene (Nostocaceae);
 Anabaena pecF gene (Nostocaceae)

L105 ANSWER 2 OF 4 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.

AN 2001:482056 BIOSIS

DN PREV200100482056

TI Biosynthesis of a fluorescent cyanobacterial C-**phycocyanin**
 holo-alpha subunit in a heterologous host.

AU Tooley, Aaron J.; Cai, Yuping A.; Glazer,
 Alexander N. (1)

CS (1) Natural Reserve System, University of California System, 1111 Franklin
 Street, 6th Floor, Oakland, CA, 94607-5200: alexander.glazer@ucop.edu USA

SO Proceedings of the National Academy of Sciences of the United States of
 America, (September 11, 2001) Vol. 98, No. 19, pp. 10560-10565. print.
 ISSN: 0027-8424.

DT Article

LA English

SL English

AB The entire pathway for the synthesis of a fluorescent

End date

holophycobiliprotein subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803) was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to the natural chromophore 3Z-**phycocyanobilin**, namely, heme oxygenase 1 and 3Z-**phycocyanobilin**:ferredoxin oxidoreductase, were expressed from a plasmid under control of the hybrid trp-lac (trc) promoter. Genes for the apoprotein (C-**phycocyanin** alpha subunit; cpcA) and the heterodimeric lyase (cpcE and cpcF) that catalyzes chromophore attachment were expressed from the trc promoter on a second plasmid. Upon induction, recombinant *E. coli* used the cellular pool of heme to produce **holo-CpcA** with spectroscopic properties qualitatively and quantitatively similar to those of the same protein produced endogenously in cyanobacteria. About a third of the apo-CpcA was converted to **holo-CpcA**. No significant **bilin** addition took place in a similarly engineered *E. coli* strain that lacks cpcE and cpcF. This approach should permit incisive analysis of many remaining questions in **phycobiliprotein** biosynthesis. These studies also demonstrate the feasibility of generating constructs of these proteins *in situ* for use as fluorescent protein probes in living cells.

- CC Genetics and Cytogenetics - General *03502
 Biochemical Studies - General *10060
 Physiology and Biochemistry of Bacteria *31000
 Genetics of Bacteria and Viruses *31500
- BC Enterobacteriaceae 06702
 Chroococcales 09210
- IT Major Concepts
 Biochemistry and Molecular Biophysics; Molecular Genetics (Biochemistry and Molecular Biophysics)
- IT Chemicals & Biochemicals
 3Z-**phycocyanobilin**: chromophore; 3Z-**phycocyanobilin**:ferredoxin oxidoreductase; C-**phycocyanin**: biosynthesis, fluorescent, **holo-alpha** subunit; heme oxygenase 1; **holophycobiliprotein**
- IT Miscellaneous Descriptors
 photosynthesis
- ORGN Super Taxa
 Chroococcales: Cyanobacteria, Oxygenic Photosynthetic Bacteria, Eubacteria, Bacteria, Microorganisms; Enterobacteriaceae: Facultatively Anaerobic Gram-Negative Rods, Eubacteria, Bacteria, Microorganisms
- ORGN Organism Name
 Escherichia coli (Enterobacteriaceae): expression system; *Synechocystis* sp. PCC6803 (Chroococcales)
- ORGN Organism Supertterms
 Bacteria; Cyanobacteria; Eubacteria; Microorganisms
- RN 93527-36-7 (3Z-**PHYCOCYANOBILIN**)
- GEN *Synechocystis* cpcA gene [*Synechocystis* C-**phycocyanin** alpha subunit gene] (Chroococcales); *Synechocystis* cpcE gene (Chroococcales); *Synechocystis* cpcF gene (Chroococcales); trp-lac hybrid gene: promoter

-
- L105 ANSWER 3 OF 4 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
- AN 1995:320261 BIOSIS
 PREV199598334561
- TI Candidate genes for the **phycoerythrocyanin** alpha subunit lyase: Biochemical analysis of pecE and pecF interposon mutants.
- AU Jung, Linda J.; Chan, Crystal F.; Glazer, Alexander N. (1)
- CS (1) MCB: Stanley/Donner ASU, 229 Stanley Hall 3206, Univ. Calif., Berkeley, CA 94720-3206 USA
- SO Journal of Biological Chemistry, (1995) Vol. 270, No. 21, pp. 12877-12884.
 ISSN: 0021-9258.
- DT Article
- LA English
- AB The rod substructures of the *Anabaena* sp. PCC 7120 **phycobilisome** contain the light harvesting proteins C-**phycocyanin** and

phycoerythrocyanin (PEC). Even at low light intensities, PEC represents no more than 5% of the **phycobilisome** protein. The beta subunits of both proteins carry thioether-linked **phycocyanobilin** (PCB) at beta-Cys-82 and beta-Cys-155; however, C-**phycocyanin** has PCB at alpha-Cys-84 whereas PEC a subunit carries **phycobiliviolin** at this position. The *Anabaena* sp. PCC 7120 pec operon is made up of five genes. PecB and pecA encode the beta and alpha subunits of PEC, pecC encodes a linker polypeptide associated with PEC in the rod substructure, and pecE and pecF are genes of unknown function that show a high degree of homology to cpcE and cpcF, that encode a C-**phycocyanin** a subunit PCB lyase (Fairchild, C. D., Zhao, J., Zhou, J., Colson, S. E., Bryant, D. A., and Glazer, A. N. (1992) Proc. Natl. Acad. Sci. U. S. A. 89, 7017-7021). Insertional mutants in pecE and pecF, and an interposon mutant in which a portion of both pecE and pecF was deleted, were constructed. All three types of mutants grew 1.3 times slower than wild-type under limiting light conditions and showed a 20% reduction in the PCB content of whole cells relative to chlorophyll alpha. **Holo-PEC** was missing from the **phycobilisomes** of all three types of mutants and the level of the PEC linker polypeptide was reduced relative to the wild-type. However, apprx 30% of the wild-type level of the PEC beta subunit was present in all of these **phycobilisomes**. In contrast, the PEC a subunit was barely detectable in the pecE and pecF mutants, but was present in the pecEF deletion mutant as a PCB-adduct in a 1:1 ratio with the PEC beta subunit. The identity of this "unnatural" adduct was confirmed by isolation of the subunit and amino-terminal sequencing. These biochemical results support the inference that pecE and pecF encode a PEC alpha subunit **phycobiliviolin** lyase, and, in conjunction with earlier findings, demonstrate that **phycobiliprotein bilin** lyases show high selectivity (rather than absolute specificity) for both the **bilin** and the polypeptide substrate.

CC Biochemical Studies - Proteins, Peptides and Amino Acids *10064
 Biochemical Studies - Porphyrins and Bile Pigments *10065
 Biophysics - Molecular Properties and Macromolecules *10506
 Enzymes - Chemical and Physical *10806
 Physiology and Biochemistry of Bacteria *31000
 Genetics of Bacteria and Viruses *31500

BC Nostocaceae *09241

IT Major Concepts

Biochemistry and Molecular Biophysics; Enzymology (Biochemistry and Molecular Biophysics); Genetics; Physiology

IT Chemicals & Biochemicals
 LYASE; LYASES

IT Miscellaneous Descriptors

PHYCOBILIPROTEIN BILIN LYASES;
 PHYCOBILISOMES

ORGN Super Taxa

Nostocaceae: Cyanobacteria, Eubacteria, Bacteria

ORGN Organism Name

Anabaena (Nostocaceae)

ORGN Organism Superterms

bacteria; cyanobacteria; eubacteria; microorganisms

RN 9055-04-3 (LYASE)

9055-04-3D (LYASES)

L105 ANSWER 4 OF 4 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.

AN 1992:506225 BIOSIS

DN BA94:124750

TI PHYCOCYANIN ALPHA-SUBUNIT PHYCOCYANOBILIN LYASE.

AU FAIRCHILD C D; ZHAO J; COLSON S E; BRYANT D A; GLAZER A N

CS MCB: STANLEY/DONNER ASU, 229 STANLEY HALL, UNIV. CALIF., BERKELEY, CALIF.
 94720.

SO PROC NATL ACAD SCI U S A, (1992) 89 (15), 7017-7021.

CODEN: PNASA6. ISSN: 0027-8424.

FS BA; OLD

LA English

AB **Phycobiliproteins**, unlike other light-harvesting proteins involved in photosynthesis, bear covalently attached chromophores. The **bilin** chromophores are attached through thioether bonds to cysteine residues. The cyanobacterium *Synechococcus* sp. PCC 7002 has eight distinct **bilin** attachment sites on seven polypeptides, all of which carry the same chromophore, **phycocyanobilin**. When two genes in the **phycocyanin** operon of this organisms, *cpcE* and *cpcF*, are inactivated by insertion, together or separately, the suprising result is elimination of correct **bilin** attachment at only one site, that on the .alpha. subunit of **phycocyanin**. We have overproduced CpcE and CpcF in *Escherichia coli*. In vitro, these proteins catalyze the attachment of **phycocyanobilin** to the .alpha. subunit of **apophycocyanin** at the appropriate site, .alpha. Cys-84, to form the correct adduct. CpcE and CpcF also efficiently catalyze the reverse reaction, in which the **bilin** from **holo-.alpha.** subunit is transferred either to the **apo-.alpha.** subunit of the same C-**phycocyanin** or to the **apo-.alpha.** subunit of a heterologous C-**phycocyanin**. The forward and reverse reactions each require both CpcE and CpcF and are specific for the .alpha.-Cys-84 position. **Phycocyanobilin** is the immediate precursor of the protein-bound **bilin**.

CC Biochemical Studies - Nucleic Acids, Purines and Pyrimidines 10062
 Biochemical Studies - Proteins, Peptides and Amino Acids 10064
 Biochemical Studies - Porphyrins and Bile Pigments 10065
 Biophysics - Molecular Properties and Macromolecules 10506
 Enzymes - Chemical and Physical *10806
 Enzymes - Physiological Studies *10808
 Metabolism - Proteins, Peptides and Amino Acids *13012
 Metabolism - Porphyrins and Bile Pigments *13013
 Physiology and Biochemistry of Bacteria *31000
 Genetics of Bacteria and Viruses *31500
 Plant Physiology, Biochemistry and Biophysics - Photosynthesis 51506

BC Enterobacteriaceae 06702
 Chroococcales 09210

IT Miscellaneous Descriptors
 ESCHERICHIA-COLI SYNECHOCOCCUS CPCE GENE CPCF GENE
APOPHYCOCYANIN ALPHA SUBUNIT ATTACHMENT SITE

RN 144378-42-7 (**PHYCOCYANIN ALPHA-SUBUNIT PHYCOCYANOBILIN LYASE**)

=> d his

(FILE 'HOME' ENTERED AT 12:54:49 ON 27 MAR 2003)
 SET COST OFF

FILE 'HCAPLUS' ENTERED AT 12:55:07 ON 27 MAR 2003
 E US20030027285/PN

L1	1 S E3
	E GLAZER A/AU
L2	262 S E3,E7,E11,E13,E14
	E TOOLEY A/AU
L3	3 S E4
	E CAI Y/AU
L4	253 S E3-E18
	E CAI YU/AU
L5	30 S E3,E10
	E CAI YUPING/AU
L6	15 S E3
L7	4 S E4

E BILIPROTEIN/CT
 E E4+ALL
 L8 6658 S E7,E5+NT
 L9 2 S HOLOPHYCOCOBILIPROTEIN OR HOLO() (PHYCOBILIPROTEIN OR PHYCOBILI#

FILE 'REGISTRY' ENTERED AT 13:01:53 ON 27 MAR 2003
 L10 1 S 9059-22-7
 L11 1 S 14875-96-8

FILE 'HCAPLUS' ENTERED AT 13:02:57 ON 27 MAR 2003
 L12 9974 S L10 OR L11
 L13 2648 S HEME OXYGENASE
 L14 30884 S HEME OR PROTOHEME OR REDUCED HEMATIN OR HEM FE OR FERROHEME O
 L15 66 S L8 AND L12-L14
 L16 2 S L9 AND L15
 L17 813 S PHYCOBILIPROTEIN
 L18 6658 S L8,L9
 L19 66 S L18 AND L12-L14
 L20 6 S L19 AND (RECOMBIN? OR CONTRUCT)
 L21 126 S L2-L7 AND L8,L9,L17,L18
 L22 4 S L12-L14 AND L21
 L23 302 S BILIN
 L24 336 S ?PHYCOCYANOBILIN?
 L25 816 S ?PHYCOBILIPROTEIN?
 L26 122 S ?PHYCOERYTHROCYANIN?
 L27 12 S ?PHYCOBILIVIOLEIN?
 L28 173 S ?PHYCOERYTHROBILIN?
 L29 2074 S ?PHYCOERYTHRIN?
 L30 112 S L2-L7 AND L23-L29
 L31 4 S L12-L14 AND L30
 L32 7 S L1,L9,L16,L20,L22,L31
 L33 6 S L32 NOT PHARMACEUTICAL/TI
 SEL RN L1

FILE 'REGISTRY' ENTERED AT 13:20:58 ON 27 MAR 2003
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 L35 1 S 20298-86-6
 L36 1 S 93527-36-7
 L37 45 S C33H38N4O6/MF AND NC4/ES AND 4/NR
 L38 44 S L37 AND BILIN?
 L39 7 S L38 AND 8 12 DIPROPANOIC AND 18 ETHYL 3 ETHYLIDENE
 L40 7 S L39 AND 2 7 13 17 TETRAMETHYL 1 19 DIOXO
 L41 7 S L35,L36,L40
 L42 1 S 347401-12-1
 E PHYCOCYANOBILIN/CN
 L43 1 S E8
 E APO-PHYCOBILIPROTEIN/CN
 E PHYCOCYANIN/CN
 L44 116 S PHYCOCYANIN (L) ALPHA (L) SUBUNIT
 L45 1 S 168680-20-4
 L46 1 S 124861-40-1
 L47 1 S 18097-67-1
 L48 174 S C33H38N4O6/MF
 L49 45 S L48 AND NC4/ES AND 4/NR
 L50 3 S L49 AND BILINE 8 12 DIPROPANOIC AND 18 ETHENYL 3 ETHYLIDENE A
 L51 3 S L47,L50
 L52 1 S 347401-21-2

FILE 'HCAPLUS' ENTERED AT 13:37:26 ON 27 MAR 2003
 L53 350 S L41 OR PHYCOCYANOBILIN
 L54 8 S L42 OR PHYCOCYANOBILIN(S)FERREDOXIN(S)OXIDOREDUCTASE
 L55 8 S L45 OR PHYCOERYTHROCYANIN LYASE
 L56 12 S L46 OR PHYCOBILIVIOLEIN

L57 25 S L12-L14 AND L53
 L58 43 S L54-L57
 L59 31 S L58 AND L8
 L60 2 S L59 AND L9

FILE 'REGISTRY' ENTERED AT 13:41:42 ON 27 MAR 2003
 L61 1 S 144378-42-7

FILE 'HCAPLUS' ENTERED AT 13:42:39 ON 27 MAR 2003
 L62 6 S L61 OR PHYCOCYANIN (S) ALPHA(S) SUBUNIT (S) PHYCOCYANOBILIN LY
 L63 28 S L54-L56, L62
 L64 7 S L63 AND L12-L14
 L65 76 S HOLO? AND L8, L53-L60, L62-L64
 L66 71 S L65 AND (PD<=20010731 OR PRD<=20010731 OR AD<=20010731)
 L67 7 S L54, L55, L62 AND L66
 L68 19 S L54, L55, L62
 L69 19 S L67, L68
 L70 36 S L2-L7 AND L53-L60, L62-L63
 L71 4 S L70 AND L66
 L72 5 S L70 AND L65
 L73 5 S L71, L72
 E GENETIC ENGINEERING/CT
 E E3+ALL
 L74 79989 S E2+NT
 L75 10 S L74 AND L65
 L76 12 S L73, L75
 L77 263 S L74 AND L12-L14
 L78 8 S L77 AND L23-L29, L53-L56, L62, L63
 L79 14 S L76, L78

FILE 'HCAPLUS' ENTERED AT 13:50:25 ON 27 MAR 2003
 SEL HIT RN

FILE 'REGISTRY' ENTERED AT 13:50:40 ON 27 MAR 2003
 L80 8 S E1-E8
 L81 10 S L41, L42, L43, L45-L47, L51, L52, L61 NOT L80

FILE 'HCAPLUS' ENTERED AT 13:52:50 ON 27 MAR 2003
 L82 7 S L42, L52
 L83 1 S L82 NOT L79
 L84 1 S L83 AND L1-L33, L53-L60, L62-L79, L82-L83

FILE 'REGISTRY' ENTERED AT 13:54:24 ON 27 MAR 2003
 L85 2 S 138263-99-7 OR 347401-20-1
 L86 2 S 114-25-0 OR 18097-67-1

FILE 'HCAPLUS' ENTERED AT 13:54:36 ON 27 MAR 2003
 L87 8 S L85
 L88 753 S L86
 L89 5 S L87 AND L1-L33, L53-L60, L62-L69, L82-L84, L87, L88 NOT L79
 L90 5 S L84, L89
 L91 2 S L90 AND L88
 L92 5 S L90, L91

FILE 'HCAPLUS' ENTERED AT 13:55:52 ON 27 MAR 2003
 SEL HIT RN

FILE 'REGISTRY' ENTERED AT 13:56:10 ON 27 MAR 2003
 L93 3 S E9-E15 NOT L80, L81
 SET COST ON
 SET COST OFF

FILE 'BIOSIS' ENTERED AT 13:56:44 ON 27 MAR 2003

L94 E GLAZER A/AU
223 S E3,E5,E10,E11
E TOOLEY A/AU
E CAI Y/AU
L95 190 S E3,E12
E CAI YU/AU
L96 7 S E3
E CAI YUPING/AU
L97 4 S E3
L98 4 S E4
E TOOLEY A/AU
L99 2 S E4
L100 425 S L94-L99
L101 4 S L100 AND HOLO?
L102 140 S L100 AND ?PHYCO?
L103 64 S L100 AND ?BILIN?
L104 68 S L100 AND ?BILIPROTEIN?
L105 4 S L101 AND L102-L104

FILE 'BIOSIS' ENTERED AT 13:58:29 ON 27 MAR 2003

FILE 'BIOSIS' ENTERED AT 14:00:43 ON 27 MAR 2003

=> d his

(FILE 'HOME' ENTERED AT 12:54:49 ON 27 MAR 2003)
SET COST OFF

FILE 'HCAPLUS' ENTERED AT 12:55:07 ON 27 MAR 2003
E US20030027285/PN

L1	1 S E3 E GLAZER A/AU	Jan Delaval Reference Librarian Biotechnology & Chemical Library CM11E07 - 703-308-4498 jan.delaval@uspto.gov
L2	262 S E3,E7,E11,E13,E14 E TOOLEY A/AU	
L3	3 S E4 E CAI Y/AU	
L4	253 S E3-E18 E CAI YU/AU	
L5	30 S E3,E10 E CAI YUPING/AU	
L6	15 S E3	
L7	4 S E4 E BILIPROTEIN/CT E E4+ALL	
L8	6658 S E7,E5+NT	
L9	2 S HOLOPHYCOCOBILIPROTEIN OR HOLO() (PHYCOBILIPROTEIN OR PHYCOBILI#	

FILE 'REGISTRY' ENTERED AT 13:01:53 ON 27 MAR 2003

L10	1 S 9059-22-7
L11	1 S 14875-96-8

FILE 'HCAPLUS' ENTERED AT 13:02:57 ON 27 MAR 2003

L12	9974 S L10 OR L11
L13	2648 S HEME OXYGENASE
L14	30884 S HEME OR PROTOHEME OR REDUCED HEMATIN OR HEM FE OR FERROHEME O
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L16	2 S L9 AND L15
L17	813 S PHYCOBILIPROTEIN
L18	6658 S L8,L9
L19	66 S L18 AND L12-L14
L20	6 S L19 AND (RECOMBIN? OR CONTRUCT)
L21	126 S L2-L7 AND L8,L9,L17,L18
L22	4 S L12-L14 AND L21
L23	302 S BILIN
L24	336 S ?PHYCOCYANOBILIN?
L25	816 S ?PHYCOBILIPROTEIN?
L26	122 S ?PHYCOERYTHROCYANIN?
L27	12 S ?PHYCOBILIVIOLIN?
L28	173 S ?PHYCOERYTHROBILIN?
L29	2074 S ?PHYCOERYTHRIN?
L30	112 S L2-L7 AND L23-L29
L31	4 S L12-L14 AND L30
L32	7 S L1,L9,L16,L20,L22,L31
L33	6 S L32 NOT PHARMACEUTICAL/TI SEL RN L1

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L34	9 S E1-E9
L35	1 S 20298-86-6
L36	1 S 93527-36-7
L37	45 S C33H38N4O6/MF AND NC4/ES AND 4/NR
L38	44 S L37 AND BILIN?
L39	7 S L38 AND 8 12 DIPROPANOIC AND 18 ETHYL 3 ETHYLIDENE
L40	7 S L39 AND 2 7 13 17 TETRAMETHYL 1 19 DIOXO
L41	7 S L35,L36,L40
L42	1 S 347401-12-1

L43 E PHYCOCYANOBILIN/CN
 1 S E8
 E APO-PHYCOBILIPROTEIN/CN
 E PHYCOCYANIN/CN
L44 116 S PHYCOCYANIN (L) ALPHA (L) SUBUNIT
L45 1 S 168680-20-4
L46 1 S 124861-40-1
L47 1 S 18097-67-1
L48 174 S C33H38N4O6/MF
L49 45 S L48 AND NC4/ES AND 4/NR
L50 3 S L49 AND BILINE 8 12 DIPROPANOIC AND 18 ETHENYL 3 ETHYLIDENE A
L51 3 S L47, L50
L52 1 S 347401-21-2

FILE 'HCAPLUS' ENTERED AT 13:37:26 ON 27 MAR 2003

L53 350 S L41 OR PHYCOCYANOBILIN
L54 8 S L42 OR PHYCOCYANOBILIN(S) FERREDOXIN(S) OXIDOREDUCTASE
L55 8 S L45 OR PHYCOERYTHROCYANIN LYASE
L56 12 S L46 OR PHYCOBILIVIOLETTIN
L57 25 S L12-L14 AND L53
L58 43 S L54-L57
L59 31 S L58 AND L8
L60 2 S L59 AND L9

FILE 'REGISTRY' ENTERED AT 13:41:42 ON 27 MAR 2003

L61 1 S 144378-42-7

FILE 'HCAPLUS' ENTERED AT 13:42:39 ON 27 MAR 2003

L62 6 S L61 OR PHYCOCYANIN (S) ALPHA(S) SUBUNIT (S) PHYCOCYANOBILIN LY
L63 28 S L54-L56, L62
L64 7 S L63 AND L12-L14
L65 76 S HOLO? AND L8, L53-L60, L62-L64
L66 71 S L65 AND (PD<=20010731 OR PRD<=20010731 OR AD<=20010731)
L67 7 S L54, L55, L62 AND L66
L68 19 S L54, L55, L62
L69 19 S L67, L68
L70 36 S L2-L7 AND L53-L60, L62-L63
L71 4 S L70 AND L66
L72 5 S L70 AND L65
L73 5 S L71, L72
 E GENETIC ENGINEERING/CT
 E E3+ALL
L74 79989 S E2+NT
L75 10 S L74 AND L65
L76 12 S L73, L75
L77 263 S L74 AND L12-L14
L78 8 S L77 AND L23-L29, L53-L56, L62, L63
L79 14 S L76, L78

=> fil heapplus

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FILE COVERS 1907 - 27 Mar 2003 VOL 138 ISS 13
FILE LAST UPDATED: 26 Mar 2003 (20030326/ED)

This file contains CAS Registry Numbers for easy and accurate substance identification.

=> d 179 all tot

L79 ANSWER 1 OF 14 HCAPLUS COPYRIGHT 2003 ACS
AN 2003:97917 HCAPLUS
DN 138:148684
TI Engineering of living cells for the expression of **holo-phyco**biliprotein**-based constructs
IN Glazer, Alexander N.; Tooley, Aaron J.; Cai,
Yuping
PA USA
SO U.S. Pat. Appl. Publ., 13 pp.
CODEN: USXXCO
DT Patent
LA English
IC ICM C12P021-04
ICS C07H021-04; C12N005-06
NCL 435069600; 435320100; 435325000; 536023200; 530380000
CC 3-2 (Biochemical Genetics)
Section cross-reference(s): 10, 13
FAN.CNT 1**

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	US 2003027285	A1	20030206	US 2001-919486	20010731 <--
	WO 2003012448	A1	20030213	WO 2002-US24245	20020730 <--
	W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				

PRAI US 2001-919486 A 20010731 <--

AB Recombinant cells which express a fluorescent **holo-phyco**biliprotein** fusion protein and methods of use are described. The cells comprises a **bilin**, a recombinant **bilin reductase**, an **apo-phyco**biliprotein**** fusion protein precursor of the fusion protein comprising a corresponding **apo-phyco**biliprotein**** domain, and a recombinant **phyco**biliprotein**** domain-**bilin lyase**, which components react to form the **holo-phyco**biliprotein**** fusion protein. Also described are **holo-phyco**biliprotein**** based transcription reporter cells and assays, which cells conditionally express a heterologous-to-the-cell, fluorescent, first **holo-phyco**biliprotein**** domain.**

ST living yeast bacteria mammal cell engineering **holo-phyco**biliprotein**** prodn

IT **Phycocyanins**

RL: BCP (Biochemical process); BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation); PROC (Process)
(C-; engineering of living cells for the expression of **holo-phyco**biliprotein****-based constructs)

- IT Enzyme functional sites
(apo-**phycobiliprotein** domain and recombinant
phycobiliprotein domain-**bilin** lyase; engineering of
living cells for the expression of **holo-**
phycobiliprotein-based constructs)
- IT Fluorescence resonance energy transfer
Genetic engineering
(engineering of living cells for the expression of **holo-**
phycobiliprotein-based constructs)
- IT **Biliproteins**
RL: BCP (Biochemical process); BPN (Biosynthetic preparation); BIOL
(Biological study); PREP (Preparation); PROC (Process)
(engineering of living cells for the expression of **holo-**
phycobiliprotein-based constructs)
- IT Bile pigments
RL: BMF (Bioindustrial manufacture); BIOL (Biological study); PREP
(Preparation)
(engineering of living cells for the expression of **holo-**
phycobiliprotein-based constructs)
- IT **Phytochromes**
RL: BSU (Biological study, unclassified); BIOL (Biological study)
(engineering of living cells for the expression of **holo-**
phycobiliprotein-based constructs)
- IT Fluorometry
(for distinguishing of protein domains; engineering of living cells for
the expression of **holo-phycobiliprotein**-based
constructs)
- IT Proteins
RL: BSU (Biological study, unclassified); BIOL (Biological study)
(green fluorescent, reporter domain of the recombinant protein;
engineering of living cells for the expression of **holo-**
phycobiliprotein-based constructs)
- IT Gene, microbial
RL: BSU (Biological study, unclassified); BIOL (Biological study)
(hol; engineering of living cells for the expression of **holo-**
phycobiliprotein-based constructs)
- IT Animal cell
(mammalian, reporter cell; engineering of living cells for the
expression of **holo-phycobiliprotein**-based
constructs)
- IT Escherichia coli
Saccharomyces cerevisiae
(reporter cell; engineering of living cells for the expression of
holo-phycobiliprotein-based constructs)
- IT Synechocystis
(strain PCC6803; engineering of living cells for the expression of
holo-phycobiliprotein-based constructs)
- IT 14875-96-8, Heme
RL: BCP (Biochemical process); BIOL (Biological study); PROC (Process)
(engineering of living cells for the expression of **holo-**
phycobiliprotein-based constructs)
-
- IT 114-25-0P, Biliverdin 18097-67-1P, Phycoerythrobilin
93527-36-7P, 3(Z)-Phycocyanobilin 124861-40-1P
, Phycobiliviolin
RL: BCP (Biochemical process); BPN (Biosynthetic preparation); BIOL
(Biological study); PREP (Preparation); PROC (Process)
(engineering of living cells for the expression of **holo-**
phycobiliprotein-based constructs)
- IT 347401-12-1P
RL: BCP (Biochemical process); BPN (Biosynthetic preparation); BIOL
(Biological study); PREP (Preparation); PROC (Process)
(gene PcyA; engineering of living cells for the expression of
holo-phycobiliprotein-based constructs)

IT 347401-21-2P
 RL: BCP (Biochemical process); BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation); PROC (Process)
 (gene PebA and PebB; engineering of living cells for the expression of **holo-phycoobiliprotein**-based constructs)

IT 9059-22-7, **Heme oxygenase**
 RL: BCP (Biochemical process); BIOL (Biological study); PROC (Process)
 (gene hol, recombinant, for **bilin** formation; engineering of living cells for the expression of **holo-phycoobiliprotein**-based constructs)

IT 168680-20-4P, **Phycoerythrobilin** lyase
 RL: BCP (Biochemical process); BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation); PROC (Process)
 (heterodimeric **phycoerythrocyanin**.alpha. subunit of, gene PecF and PecE and heterodimeric C-**phycoerythrin** apo-.alpha. subunit domain of, gene CpeY and CpeZ; engineering of living cells for the expression of **holo-phycoobiliprotein**-based constructs)

L79 ANSWER 2 OF 14 HCPLUS COPYRIGHT 2003 ACS
 AN 2002:927638 HCPLUS
 DN 137:365572
 TI Use of phytochromes for light-controlled gene expression and protein translocation into nucleus
 IN Lagarius, John Clark; Kochi, Takayuki; Frankenberg, Nicole; Gambetta, Gregory A.; Montgomery, Beronda L.
 PA The Regents of the University of California, USA
 SO PCT Int. Appl., 102 pp.
 CODEN: PIXXD2
 DT Patent
 LA English
 IC ICM C12Q001-68
 ICS C12N015-63; C12N015-85; C12N015-87; C12N015-82
 CC 7-5 (Enzymes)
 Section cross-reference(s): 3, 10, 11, 16

FAN.CNT 1

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	WO 2002097137	A1	20021205	WO 2002-US17266	20020529
	W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				

PRAI US 2001-294463P P 20010529

AB This invention relates to the use of heterologous phytochromes to translocate polypeptides into the nucleus of a cell. Where the polypeptides comprise transactivators or repressors this invention provides a system for light-directed gene expression. This invention identifies a novel family of **bilin** reductases. Designated herein HY2 **bilin** reductases, the enzymes of this invention are useful in a wide variety of contexts including but not limited to the conversion of biliverdins to phytobilins and the assembly of **holophytochromes** or phytofluors. The HY2 family of **bilin** reductases are ferredoxin-dependent. Using the HY2 protein sequence as a query sequence, HY2 family members were identified in the genomes of various cyanobacteria, oxyphotobacteria and plants.

ST phytochrome apoprotein light induction gene expression protein transport nucleus; HY2 **bilin** reductase cyanobacteria oxyphotobacteria

plant; ferredoxin dependent **bilin** reductase HY2; phytobilin phytofluor fermn HY2 **bilin** reductase
IT Nuclear receptors
RL: BSU (Biological study, unclassified); BIOL (Biological study)
(DNA binding domain of Gal4 transcription factor; use of phytochromes for light-controlled gene expression and protein translocation into nucleus)

IT Transcription factors
RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(GAL4, as transactivator for gene expression; use of phytochromes for light-controlled gene expression and protein translocation into nucleus)

IT Gene
RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(HO1; cloning and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT Gene, plant
RL: BSU (Biological study, unclassified); BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(HY2; cloning and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT Transcription factors
RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(NF-I (nuclear factor I), as transactivator for gene expression; use of phytochromes for light-controlled gene expression and protein translocation into nucleus)

IT Transcription factors
RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(Spl, as transactivator for gene expression; use of phytochromes for light-controlled gene expression and protein translocation into nucleus)

IT Transcription factors
RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(VP16, as transactivator for gene expression; use of phytochromes for light-controlled gene expression and protein translocation into nucleus)

IT Proteins
RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(apoproteins, phytochrome; use of phytochromes for light-controlled gene expression and protein translocation into nucleus)

IT Anabaena
Arabidopsis thaliana
Barley
Cyanobacteria
Embryophyta
Fermentation
Fluorescent indicators
Molecular cloning
Plastid
Prochlorococcus marinus
Synechococcus
Synechocystis
(cloning and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT Fusion proteins (chimeric proteins)
RL: BCP (Biochemical process); BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation); PROC (Process)

- (cloning and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)
- IT Phytochromes
 RL: BMF (Bioindustrial manufacture); BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation)
 (cloning and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)
- IT Transcription factors
 RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
 (lactose repressors, for gene expression; use of phytochromes for light-controlled gene expression and protein translocation into nucleus)
- IT Proteins
 RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
 (linker, peptide; use of phytochromes for light-controlled gene expression and protein translocation into nucleus)
- IT Evolution
 (mol., of HY2 family of **bilin** reductases; cloning and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)
- IT Gene, microbial
 RL: BSU (Biological study, unclassified); BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
 (pcyA; cloning and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)
- IT Operon
 (peb; cloning and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)
- IT Gene, microbial
 RL: BSU (Biological study, unclassified); BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
 (pebA; cloning and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)
- IT Gene, microbial
 RL: BSU (Biological study, unclassified); BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
 (pebB; cloning and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)
- IT Bile pigments
 RL: BMF (Bioindustrial manufacture); BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation)
 (phytobilins; cloning and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)
- IT Bile pigments
 RL: BMF (Bioindustrial manufacture); BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation)
 (phytofluors; cloning and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)
-
- IT Cytoplasm
 (protein targeting to nucleus from; use of phytochromes for light-controlled gene expression and protein translocation into nucleus)
- IT Transcription factors
 RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
 (repressors, lex, for gene expression; use of phytochromes for light-controlled gene expression and protein translocation into nucleus)
- IT Transcription factors
 RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)

(repressors, tet, for gene expression; use of phytochromes for light-controlled gene expression and protein translocation into nucleus)

IT Biological transport
 Cell nucleus
 Genome
 Light
 (use of phytochromes for light-controlled gene expression and protein translocation into nucleus)

IT Proteins
 RL: BPN (Biosynthetic preparation); BSU (Biological study, unclassified); BIOL (Biological study); PREP (Preparation)
 (use of phytochromes for light-controlled gene expression and protein translocation into nucleus)

IT 114-25-0, Biliverdin 78249-71-5, Phytochromobilin
 RL: BCP (Biochemical process); BIOL (Biological study); PROC (Process)
 (cloning and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT 9059-22-7P, Heme oxygenase
 RL: BCP (Biochemical process); BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation); PROC (Process)
 (cloning and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT 160047-82-5P, Biliverdin IX.alpha. reductase 199618-44-5P
 RL: BCP (Biochemical process); BPN (Biosynthetic preparation); BSU (Biological study, unclassified); BIOL (Biological study); PREP (Preparation); PROC (Process)
 (cloning and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT 138263-99-7P
 RL: BCP (Biochemical process); BPN (Biosynthetic preparation); BSU (Biological study, unclassified); BIOL (Biological study); PREP (Preparation); PROC (Process)
 (gene HY2; cloning and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT 347401-12-1P
 RL: BCP (Biochemical process); BPN (Biosynthetic preparation); BSU (Biological study, unclassified); BIOL (Biological study); PREP (Preparation); PROC (Process)
 (gene pcyA; cloning and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT 347401-20-1P
 RL: BCP (Biochemical process); BPN (Biosynthetic preparation); BSU (Biological study, unclassified); BIOL (Biological study); PREP (Preparation); PROC (Process)
 (gene pebA; cloning and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT 347401-21-2P
 RL: BCP (Biochemical process); BPN (Biosynthetic preparation); BSU (Biological study, unclassified); BIOL (Biological study); PREP (Preparation); PROC (Process)
 (gene pebB; cloning and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

RE.CNT 4 THERE ARE 4 CITED REFERENCES AVAILABLE FOR THIS RECORD

- RE
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 - (3) Quail; US 5656496 A 1997 HCPLUS
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L79 ANSWER 3 OF 14 HCPLUS COPYRIGHT 2003 ACS

AN 2002:629384 HCPLUS

DN 138:181625

TI Biosynthesis of the cyanobacterial light-harvesting polypeptide
phycoerythrocyanin holo-.alpha. subunit in a
 heterologous host

AU Tooley, Aaron J.; Glazer, Alexander N.

CS Department of Molecular and Cell Biology, University of California,
 Berkeley, CA, 94720-3200, USA

SO Journal of Bacteriology (2002), 184(17), 4666-4671
 CODEN: JOBAAY; ISSN: 0021-9193

PB American Society for Microbiology

DT Journal

LA English

CC 3-2 (Biochemical Genetics)
 Section cross-reference(s): 6, 10

AB The entire pathway for the biosynthesis of the **phycobiliviolin**-bearing His-tagged **holo-.alpha.** subunit of the cyanobacterial photosynthetic accessory protein **phycoerythrocyanin** was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of **heme** to 3Z-**phycocyanobilin**, a precursor of **phycobiliviolin** (namely, **heme oxygenase 1** and 3Z-**phycocyanobilin ferredoxin oxidoreductase**), were expressed from a plasmid under the control of the hybrid trp-lac (trc) promoter. Genes for the apo-**phycoerythrocyanin .alpha.** subunit (pecA) and the heterodimeric lyase/isomerase (pecE and pecF), which catalyzes both the covalent attachment of **phycocyanobilin** and its concurrent isomerization to **phycobiliviolin**, were expressed from the trc promoter on a second plasmid. Upon induction, recombinant *E. coli* used endogenous **heme** to produce **holo-PecA** with absorbance and fluorescence properties similar to those of the same protein produced in cyanobacteria. About two-thirds of the apo-PecA was converted to **holo-PecA**. No significant **bilin** addn. took place in a similarly engineered *E. coli* strain that lacks pecE and pecF. By using immobilized metal affinity chromatog., both apo-PecA and **holo-PecA** were isolated as ternary complexes with PecE and PecF. The identities of all three components in the ternary complexes were established unambiguously by protein and tryptic peptide analyses performed by matrix-assisted laser desorption ionization-time of flight mass spectrometry.

ST Anabaena cyanobacteria **phycoerythrocyanin pecA holo alpha** subunit *Escherichia*

IT **Genetic engineering**
 (Biosynthesis of the cyanobacterial light-harvesting polypeptide **phycoerythrocyanin holo-.alpha.** subunit in a heterologous host)

IT Anabaena
Escherichia coli
 (biosynthesis of Anabaena light-harvesting polypeptide **phycoerythrocyanin holo-.alpha.** subunit in *Escherichia coli*)

IT Gene, microbial
 RL: BSU (Biological study, unclassified); BIOL (Biological study)
 (pecA; biosynthesis of Anabaena light-harvesting polypeptide **phycoerythrocyanin holo-.alpha.** subunit in *Escherichia coli*)

IT Gene, microbial
 RL: BSU (Biological study, unclassified); BIOL (Biological study)
 (pecE; biosynthesis of Anabaena light-harvesting polypeptide **phycoerythrocyanin holo-.alpha.** subunit in *Escherichia coli*)

IT Gene, microbial
 RL: BSU (Biological study, unclassified); BIOL (Biological study)
 (pecF; biosynthesis of Anabaena light-harvesting polypeptide **phycoerythrocyanin holo-.alpha.** subunit in *Escherichia coli*)

bad date

- IT Escherichia coli)
- IT **Phycocyanins**
Phycoerythrins
 RL: BPN (Biosynthetic preparation); BSU (Biological study, unclassified); BIOL (Biological study); PREP (Preparation)
(phycoerythrocyanins, .alpha. subunit, complex with PecE and PecF; biosynthesis of Anabaena light-harvesting polypeptide phycoerythrocyanin holo-.alpha. subunit in Escherichia coli)
- IT **9059-22-7**
 RL: BSU (Biological study, unclassified); BIOL (Biological study)
(1, gene for; biosynthesis of Anabaena light-harvesting polypeptide phycoerythrocyanin holo-.alpha. subunit in Escherichia coli in relation to)
- IT **14875-96-8, Heme 20298-86-6,**
Phycocyanobilin 124861-40-1, Phycobiliviolin
 RL: BSU (Biological study, unclassified); BIOL (Biological study)
(biosynthesis of Anabaena light-harvesting polypeptide phycoerythrocyanin holo-.alpha. subunit in Escherichia coli)
- IT **347401-12-1, 3Z-Phycocyanobilin:ferredoxin oxidoreductase**
 RL: BSU (Biological study, unclassified); BIOL (Biological study)
(gene for; biosynthesis of Anabaena light-harvesting polypeptide phycoerythrocyanin holo-.alpha. subunit in Escherichia coli in relation to)
- IT **168680-20-4, Phycoerythrocyanin .alpha. subunit lyase**
 RL: BSU (Biological study, unclassified); BIOL (Biological study)
(genes pecE and pecF for; biosynthesis of Anabaena light-harvesting polypeptide phycoerythrocyanin holo-.alpha. subunit in Escherichia coli)

RE.CNT 22 THERE ARE 22 CITED REFERENCES AVAILABLE FOR THIS RECORD

- RE
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 - (3) Bishop, J; J Am Chem Soc 1987, V10, P875
 - (4) Bryant, D; Arch Microbiol 1976, V110, P60
 - (5) Bryant, D; J Gen Microbiol 1982, V128, P835 HCPLUS
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<http://www3.interscience.wiley.com> 1998
 - (15) Jung, L; J Biol Chem 1995, V270, P12877 HCPLUS
 - (16) Kaneko, T; DNA Res 2001, V31, P205
 - (17) Laemmli, U; Nature 1970, V227, P680 HCPLUS
 - (18) Nakamura, Y; Nucleic Acids Res 2000, V28, P72 HCPLUS
 - (19) Storf, M; Biochemistry 2001, V40, P12444 HCPLUS
 - (20) Tooley, A; Proc Natl Acad Sci 2001, V98, P10560 HCPLUS
 - (21) Williams, V; J Biol Chem 1978, V253, P202 HCPLUS
 - (22) Zhao, K; FEBS Lett 2000, V469, P9 HCPLUS

L79 ANSWER 4 OF 14 HCPLUS COPYRIGHT 2003 ACS

AN 2001:904470 HCPLUS

DN 136:50278

TI Identification, cloning, sequences and use of HY2 family of ferredoxin-dependent bilin reductases from bacteria and plants

IN Lagarias, John Clark; Rochi, Takayuki; Frankenberg, Nicole; Gambetta,

PA Gregory A.; Montgomery, Beronda L.
 SO The Regents of the University of California, USA
 PCT Int. Appl., 102 pp.
 CODEN: PIXXD2

DT Patent
 LA English
 ICI C12
 CC 7-5 (Enzymes)
 Section cross-reference(s): 3, 10, 11, 16

FAN.CNT 1

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	WO 2001094548	A2	20011213	WO 2001-US18326	20010605
	WO 2001094548	A3	20020711		
	W: CA, JP				
	RW: AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR				
	EP 1290135	A2	20030312	EP 2001-942007	20010605
	R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI, CY, TR				
PRAI	US 2000-210286P	P	20000608		
	WO 2001-US18326	W	20010605		
AB	This invention identifies a novel family of bilin reductases. Designated herein HY2 bilin reductases, the enzymes of this invention are useful in a wide variety of contexts including but not limited to the conversion of biliverdins to phytobilins and the assembly of holophytochromes or phytofluors. The HY2 family of bilin reductases are ferredoxin-dependent. The genomic sequence and the encoded protein sequence of the gene HY2 phytochromobilin synthase of <i>Arabidopsis thaliana</i> are disclosed. Using the HY2 protein sequence as a query sequence, HY2 family members were identified in the genomes of various cyanobacteria, oxyphotobacteria and plants.				
ST	HY2 bilin reductase cyanobacteria oxyphotobacteria plant sequence; ferredoxin dependent bilin reductase HY2 sequence; phytobilin phytofluor fermn HY2 bilin reductase				
IT	Gene RL: BUU (Biological use, unclassified); PRP (Properties); BIOL (Biological study); USES (Uses) (HO1; identification, cloning, sequences and use of HY2 family of ferredoxin-dependent bilin reductases from bacteria and plants)				
IT	Gene, plant RL: BSU (Biological study, unclassified); BUU (Biological use, unclassified); PRP (Properties); BIOL (Biological study); USES (Uses) (HY2; identification, cloning, sequences and use of HY2 family of ferredoxin-dependent bilin reductases from bacteria and plants)				
IT	Bacteria (Eubacteria) Insecta Plant cell Yeast (cloning host; identification, cloning, sequences and use of HY2 family of ferredoxin-dependent bilin reductases from bacteria and plants)				
IT	Algae Anabaena <i>Arabidopsis thaliana</i> Barley Cyanobacteria DNA sequences Embryophyta Fermentation Fluorescent indicators				

Molecular cloning

Nostoc punctiforme

Oxyphotobacteria

Plastid

Prochlorococcus

Protein sequences

Synechococcus

Synechocystis

(identification, cloning, sequences and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT Fusion proteins (chimeric proteins)

RL: BCP (Biochemical process); BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation); PROC (Process)

(identification, cloning, sequences and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)IT **Phytochromes**

RL: BMF (Bioindustrial manufacture); BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation)

(identification, cloning, sequences and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT Animal cell

(mammalian, cloning host; identification, cloning, sequences and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT Evolution

(mol.; identification, cloning, sequences and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT Gene, microbial

RL: BSU (Biological study, unclassified); BUU (Biological use, unclassified); PRP (Properties); BIOL (Biological study); USES (Uses) (pcyA; identification, cloning, sequences and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT Operon

(peb; identification, cloning, sequences and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT Gene, microbial

RL: BSU (Biological study, unclassified); BUU (Biological use, unclassified); PRP (Properties); BIOL (Biological study); USES (Uses) (pebA; identification, cloning, sequences and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT Gene, microbial

RL: BSU (Biological study, unclassified); BUU (Biological use, unclassified); PRP (Properties); BIOL (Biological study); USES (Uses) (pebB; identification, cloning, sequences and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT Bile pigments

RL: BMF (Bioindustrial manufacture); BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation) (phytobilins; identification, cloning, sequences and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT Bile pigments

RL: BMF (Bioindustrial manufacture); BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation) (phytofluors; identification, cloning, sequences and use of HY2 family

of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT 381665-56-1P 381665-57-2P 381665-58-3P 381665-59-4P 381665-60-7P
 381665-84-5P 381665-85-6P 381665-86-7P 381665-87-8P 381665-88-9P
 381665-89-0P 381665-90-3P 381665-91-4P 381665-92-5P 381665-93-6P
 381665-94-7P 381665-95-8P 381665-96-9P 381665-97-0P 381665-98-1P
 381665-99-2P 381666-00-8P 381741-62-4P

RL: BCP (Biochemical process); BPN (Biosynthetic preparation); BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study); PREP (Preparation); PROC (Process)
 (amino acid sequence; identification, cloning, sequences and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT 138263-99-7P
 RL: BCP (Biochemical process); BPN (Biosynthetic preparation); BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study); PREP (Preparation); PROC (Process)
 (gene HY2; identification, cloning, sequences and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT 347401-12-1P
 RL: BCP (Biochemical process); BPN (Biosynthetic preparation); BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study); PREP (Preparation); PROC (Process)
 (gene pcyA; identification, cloning, sequences and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT 347401-20-1P
 RL: BCP (Biochemical process); BPN (Biosynthetic preparation); BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study); PREP (Preparation); PROC (Process)
 (gene pebA; identification, cloning, sequences and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT 347401-21-2P
 RL: BCP (Biochemical process); BPN (Biosynthetic preparation); BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study); PREP (Preparation); PROC (Process)
 (gene pebB; identification, cloning, sequences and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT 114-25-0, Biliverdin 78249-71-5, Phytochromobilin
 RL: BCP (Biochemical process); BIOL (Biological study); PROC (Process)
 (identification, cloning, sequences and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT 9059-22-7P, Heme oxygenase
 RL: BCP (Biochemical process); BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation); PROC (Process)
 (identification, cloning, sequences and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT 160047-82-5P, Biliverdin IX.alpha. reductase 199618-44-5P
 RL: BCP (Biochemical process); BPN (Biosynthetic preparation); BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study); PREP (Preparation); PROC (Process)
 (identification, cloning, sequences and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT 381741-61-3
 RL: BCP (Biochemical process); BSU (Biological study, unclassified); BUU (Biological use, unclassified); PRP (Properties); BIOL (Biological study); PROC (Process); USES (Uses)

(nucleotide sequence; identification, cloning, sequences and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

IT 381747-43-9 381747-44-0 381747-45-1 381747-46-2 381747-47-3
 381747-48-4 381747-49-5 381747-50-8 381747-51-9 381747-52-0
 381747-53-1 381747-54-2 381747-55-3 381747-56-4 381747-57-5
 381747-58-6 381747-59-7 381747-60-0 381747-61-1 381747-62-2
 381747-63-3 381747-64-4 381747-65-5 381747-66-6 381747-67-7
 381747-68-8 381747-69-9 381747-70-2 381747-71-3 381747-72-4
 381747-74-6

RL: PRP (Properties)

(unclaimed nucleotide sequence; identification, cloning, sequences and use of HY2 family of ferredoxin-dependent **bilin** reductases from bacteria and plants)

L79 ANSWER 5 OF 14 HCAPLUS COPYRIGHT 2003 ACS

AN 2001:866304 HCAPLUS

DN 136:180441

TI Recombinant **holophytochrome** in Escherichia coli

AU Landgraf, F. T.; Forreiter, C.; Hurtado Pico, A.; Lamarter, T.; Hughes, J.

CS Plant Physiology, Daz Zeughaus, Justus-Liebig-University Giessen, Giessen, D-35390, Germany

SO FEBS Letters (2001), 508(3), 459-462

CODEN: FEBLAL; ISSN: 0014-5793

PB Elsevier Science B.V.

DT Journal

LA English

CC 10-4 (Microbial, Algal, and Fungal Biochemistry)

AB We have successfully co-expressed two genes from the **bilin**

biosynthetic pathway of Synechocystis together with cyanobacterial

phytochrome 1 (Cph1) from the same organism to produce

holophytochrome in Escherichia coli. **Heme**

oxygenase was used to convert host **heme** to biliverdin

IX.alpha. which was then reduced to **phycocyanobilin** via

phycocyanobilin:ferredoxin oxidoreductase,

presumably with the aid of host **ferredoxin**. In this host

environment Cph1 apophytochrome was able to autoassemble with the

phycocyanobilin in vivo to form fully photoreversible

holophytochrome. The system can be used as a tool for further

genetic studies of phytochrome function and signal transduction as well as

providing an excellent source of **holophytochrome** for

physicochem. studies.

ST Escherichia recombinant **holophytochrome** formation

IT **Phytochromes**

RL: BSU (Biological study, unclassified); BIOL (Biological study)

(Phytochrome 1, apo- and **holo-**; recombinant

holophytochrome in Escherichia coli)

IT Gene, microbial

RL: BSU (Biological study, unclassified); BIOL (Biological study)
 (cph1; recombinant **holophytochrome** in Escherichia coli)

IT Gene, microbial

RL: BSU (Biological study, unclassified); BIOL (Biological study)
 (hol; recombinant **holophytochrome** in Escherichia coli)

IT Gene, microbial

RL: BSU (Biological study, unclassified); BIOL (Biological study)
 (pcyA; recombinant **holophytochrome** in Escherichia coli)

IT **Molecular cloning**

Synechocystis

(recombinant **holophytochrome** in Escherichia coli)

IT Ferredoxins

RL: BSU (Biological study, unclassified); BIOL (Biological study)
 (recombinant **holophytochrome** in Escherichia coli)

IT Escherichia coli
 (recombinant; recombinant **holophytochrome** in Escherichia coli)
 IT 114-25-0, Biliverdin IX.alpha. 9059-22-7, **Heme oxygenase** 14875-96-8, **Heme** 20298-86-6, **Phycocyanobilin** 347401-12-1
 RL: BSU (Biological study, unclassified); BIOL (Biological study)
 (recombinant **holophytochrome** in Escherichia coli)

RE.CNT 17 . THERE ARE 17 CITED REFERENCES AVAILABLE FOR THIS RECORD
 RE

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L79 ANSWER 6 OF 14 HCPLUS COPYRIGHT 2003 ACS

AN 2001:705482 HCPLUS

DN 135:369332

TI Genetic engineering of phytochrome biosynthesis in bacteria

AU Gambetta, Gregory A.; Lagarias, J. Clark

CS Section of Molecular and Cellular Biology, University of California, Davis, CA, 95616, USA

SO Proceedings of the National Academy of Sciences of the United States of America (2001), 98(19), 10566-10571

CODEN: PNASA6; ISSN: 0027-8424

PB National Academy of Sciences

DT Journal

LA English

CC 11-8 (Plant Biochemistry)

Section cross-reference(s): 3

AB The **bilin** prosthetic groups of the phytochrome photoreceptors and the light-harvesting **phycobiliprotein** antennae arise from the oxygen-dependent ring opening of **heme**. Two ferredoxin-dependent enzymes contribute to this conversion: a **heme oxygenase** and a **bilin** reductase with discrete double-bond specificity. Using a dual plasmid system, one expressing a truncated-cyanobacterial apophytochrome 1, Cph1(N514), and the other expressing a two-gene operon consisting of a **heme oxygenase** and a **bilin** reductase, these studies establish the feasibility of producing photoactive phytochromes in any **heme**-contg. cell. Heterologous expression systems for phytochromes not only will facilitate genetic anal. of their assembly, spectrophotometric activity, and biol. function, but also might afford the means to regulate gene expression by light in nonplant cells.

ST Escherichia phytochrome biosynthesis

IT Proteins, specific or class

RL: BPN (Biosynthetic preparation); PUR (Purification or recovery); BIOL (Biological study); PREP (Preparation)
 (Cph1(N514); genetic engineering of phytochrome biosynthesis in bacteria)

IT Promoter (genetic element)
 RL: BPR (Biological process); BSU (Biological study, unclassified); BIOL (Biological study); PROC (Process)
 (ara; genetic engineering of phytochrome biosynthesis in bacteria)

IT Gene, microbial
 RL: BPR (Biological process); BSU (Biological study, unclassified); BIOL (Biological study); PROC (Process)
 (cphl; genetic engineering of phytochrome biosynthesis in bacteria)

IT Molecular cloning
 (genetic engineering of phytochrome biosynthesis in bacteria)

IT Phytochromes
 RL: BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation)
 (genetic engineering of phytochrome biosynthesis in bacteria)

IT Gene, microbial
 RL: BPR (Biological process); BSU (Biological study, unclassified); BIOL (Biological study); PROC (Process)
 (hol; genetic engineering of phytochrome biosynthesis in bacteria)

IT Gene, microbial
 RL: BPR (Biological process); BSU (Biological study, unclassified); BIOL (Biological study); PROC (Process)
 (pcyA; genetic engineering of phytochrome biosynthesis in bacteria)

IT Escherichia coli
 (recombrnant; genetic engineering of phytochrome biosynthesis in bacteria)

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L79 ANSWER 7 OF 14 HCAPLUS COPYRIGHT 2003 ACS
 AN 2001:705481 HCAPLUS
 DN 136:2664
 TI Biosynthesis of a fluorescent cyanobacterial C-phycocyanin **holo**-alpha. subunit in a heterologous host
 AU Tooley, Aaron J.; Cai, Yuping A.; Glazer, Alexander N.
 CS Department of Molecular and Cell Biology, University of California, Berkeley, CA, 94720-3200, USA
 SO Proceedings of the National Academy of Sciences of the United States of America (2001), 98(19), 10560-10565
 CODEN: PNASA6; ISSN: 0027-8424
 PB National Academy of Sciences
 DT Journal
 LA English
 CC 10-2 (Microbial, Algal, and Fungal Biochemistry)
 Section cross-reference(s): 3
 AB The entire pathway for the synthesis of a fluorescent **holophycobiliprotein** subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803) was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of **heme** to the natural chromophore 3Z-**phycocyanobilin**, namely, **heme oxygenase** 1 and 3Z-**phycocyanobilin:ferredoxin oxidoreductase**, were expressed from a plasmid under control of the hybrid trp-lac (trc) promoter. Genes for the apoprotein (C-phycocyanin .alpha. subunit; cpcA) and the heterodimeric lyase (cpcE and cpcF) that catalyzes chromophore attachment were expressed from the trc promoter on a second plasmid. Upon induction, recombinant *E. coli* used the cellular pool of **heme** to produce **holo**-CpcA with spectroscopic properties qual. and quant. similar to those of the same protein produced endogenously in cyanobacteria. About a third of the apo-CpcA was converted to **holo**-CpcA. No significant **bilin** addn. took place in a similarly engineered *E. coli* strain that lacks cpcE and cpcF. This approach should permit incisive anal. of many remaining questions in **phycobiliprotein** biosynthesis. These studies also demonstrate the feasibility of generating constructs of these proteins *in situ* for use as fluorescent protein probes in living cells.
 ST *Synechocystis* phycocyanin formation
 IT **Phycocyanins**
 RL: BSU (Biological study, unclassified); MFM (Metabolic formation); BIOL (Biological study); FORM (Formation, nonpreparative)
 (C-; biosynthesis of fluorescent cyanobacterial C-phycocyanin **holo**-alpha. subunit in heterologous host)
 IT *Escherichia coli*
 Molecular cloning
Synechocystis
 (biosynthesis of fluorescent cyanobacterial C-phycocyanin **holo**-alpha. subunit in heterologous host)
 IT 144378-42-7P, Phycocyanin .alpha.-subunit phycocyanobilin lyase
 347401-12-1P, Ferredoxin:3Z-phycocyanobilin oxidoreductase
 RL: BAC (Biological activity or effector, except adverse); BPN (Biosynthetic preparation); BSU (Biological study, unclassified); BIOL (Biological study); PREP (Preparation)
 (biosynthesis of fluorescent cyanobacterial C-phycocyanin **holo**-alpha. subunit in heterologous host)
 IT 14875-96-8, Heme
 RL: BPR (Biological process); BSU (Biological study, unclassified); BIOL (Biological study); PROC (Process)
 (biosynthesis of fluorescent cyanobacterial C-phycocyanin **holo**-alpha. subunit in heterologous host)

IT -.alpha. subunit in heterologous host)
93527-36-7
 RL: BPR (Biological process); BSU (Biological study, unclassified); MFM (Metabolic formation); BIOL (Biological study); FORM (Formation, nonpreparative); PROC (Process)
 (biosynthesis of fluorescent cyanobacterial C-phycocyanin **holo** -.alpha. subunit in heterologous host)

IT **9059-22-7P, Heme oxygenase**
 RL: BAC (Biological activity or effector, except adverse); BPN (Biosynthetic preparation); BSU (Biological study, unclassified); BIOL (Biological study); PREP (Preparation)
 (isoform 1; biosynthesis of fluorescent cyanobacterial C-phycocyanin **holo**-.alpha. subunit in heterologous host)

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L79 ANSWER 8 OF 14 HCPLUS COPYRIGHT 2003 ACS

AN 2001:465506 HCPLUS

DN 135:192925

TI The heme-oxygenase family required for phytochrome chromophore biosynthesis is necessary for proper photomorphogenesis in higher plants

AU Davis, Seth J.; Bhoo, Seong Hee; Durski, Adam M.; Walker, Joseph M.; Vierstra, Richard D.

CS Laboratory of Genetics, Cellular and Molecular Biology Program, University of Wisconsin, Madison, WI, 53706, USA

SO Plant Physiology (2001), 126(2), 656-669
 CODEN: PLPHAY; ISSN: 0032-0889

PB American Society of Plant Physiologists

DT Journal

LA English

CC 11-3 (Plant Biochemistry)
 Section cross-reference(s): 3

AB The committed step in the biosynthesis of the phytochrome chromophore

phytochromobilin involves the oxidative cleavage of heme by a heme oxygenase (HO) to form biliverdin IX.alpha.. Through positional cloning of the photomorphogenic mutant *hyl*, the *Arabidopsis* HO (designated AtHO1) responsible for much of phytochromobilin synthesis recently was identified. Using the AtHO1 sequence, we identified families of HO genes in a no. of plants that cluster into two subfamilies (HO1- and HO2-like). The tomato (*Lycopersicon esculentum*) *yg-2* and *Nicotiana plumbaginifolia* *pew1* photomorphogenic mutants are defective in specific HO genes. Phenotypic anal. of a T-DNA insertion mutant of *Arabidopsis* HO2 revealed that the second HO subfamily also contributes to phytochromobilin synthesis. Homozygous *ho2-1* plants show decreased chlorophyll accumulation, reduced growth rate, accelerated flowering time, and reduced de-etiolation. A mixt. of apo- and **holo**-phyA was detected in etiolated *ho2-1* seedlings, suggesting that phytochromobilin is limiting in this mutant, even in the presence of functional AtHO1. The patterns of *Arabidopsis* HO1 and HO2 expression suggest that the products of both genes overlap temporally and spatially. Taken together, the family of HOs is important for phytochrome-mediated development in a no. of plants and that each family member may uniquely contribute to the phytochromobilin pool needed to assemble **holo**-phytochromes.

- ST *Arabidopsis* HO gene heme oxygenase phytochrome mol cloning
- IT Gene, plant
 RL: BOC (Biological occurrence); BSU (Biological study, unclassified);
 BIOL (Biological study); OCCU (Occurrence)
 (atho1; heme-oxygenase family is necessary for proper
 photomorphogenesis in higher plants)
- IT Gene, plant
 RL: BOC (Biological occurrence); BSU (Biological study, unclassified);
 BIOL (Biological study); OCCU (Occurrence)
 (atho2; heme-oxygenase family is necessary for proper
 photomorphogenesis in higher plants)
- IT *Arabidopsis thaliana*
 Molecular cloning
 Tobacco (*Nicotiana plumbaginifolia*)
 Tomato
 Transformation, genetic
 (heme-oxygenase family is necessary for proper photomorphogenesis in
 higher plants)
- IT **Phytochromes**
 RL: BPR (Biological process); BSU (Biological study, unclassified); BIOL
 (Biological study); PROC (Process)
 (heme-oxygenase family is necessary for proper photomorphogenesis in
 higher plants)
- IT Protein sequences
 (of heme oxygenase; heme-oxygenase family is necessary for proper
 photomorphogenesis in higher plants)
- IT Growth and development, plant
 (photomorphogenesis; heme-oxygenase family is necessary for proper
 photomorphogenesis in higher plants)
- IT Proteins, specific or class
 RL: BOC (Biological occurrence); BSU (Biological study, unclassified);
 BIOL (Biological study); OCCU (Occurrence)
 (phy; heme-oxygenase family is necessary for proper photomorphogenesis
 in higher plants)
- IT 9059-22-7, Heme oxygenase
 RL: BAC (Biological activity or effector, except adverse); BSU (Biological
 study, unclassified); PRP (Properties); BIOL (Biological study)
 (heme-oxygenase family is necessary for proper photomorphogenesis in
 higher plants)
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L79 ANSWER 9 OF 14 HCPLUS COPYRIGHT 2003 ACS
 AN 2000:584024 HCPLUS
 DN 134:25870
 TI Phytobilin biosynthesis: the *Synechocystis* sp. PCC 6803 **heme oxygenase**-encoding *hol* gene complements a phytochrome-deficient *Arabidopsis thaliana* *hy1* mutant
 AU Willows, Robert D.; Mayer, Sandra M.; Foulk, Michael S.; DeLong, Alison; Hanson, Kimberly; Chory, Joanne; Beale, Samuel I.
 CS Division of Biology and Medicine, Brown University, Providence, RI, 02912, USA
 SO Plant Molecular Biology (2000), 43(1), 113-120
 CODEN: PMBIDB; ISSN: 0167-4412
 PB Kluwer Academic Publishers

DT Journal
 LA English
 CC 3-2 (Biochemical Genetics)
 Section cross-reference(s): 7, 11
 AB The phytobilin chromophores of **phytocobiliproteins** and phytochromes are biosynthesized from **heme** in a pathway that begins with the opening of the tetrapyrrole macrocycle of **protoheme** to form biliverdin IX. α , in a reaction catalyzed by **heme oxygenase**. An *Arabidopsis thaliana* hyl mutant was previously shown to be deficient in phytochrome responses, and these responses were regained when the plants were administered biliverdin IX. α . A **heme oxygenase**-encoding gene, hol, was recently cloned from the cyanobacterium *Synechocystis* sp. PCC 6083. When hol was expressed in *Escherichia coli*, the cells produced active ferredoxin-dependent sol. **heme oxygenase**. The open reading frame of hol was fused in frame with a chloroplast transit peptide-encoding sequence from the oli gene of *Antirrhinum majus*. This construct was placed in a binary plasmid vector contg. a kanamycin resistance marker and a cauliflower mosaic virus 35S promoter to control expression of the chimeric oli-hol gene and used to transform *A. thaliana* hyl plants. Two independent transformed lines were obtained that had the phenotype of the parental Landsberg erecta line and expressed the chimeric gene, as indicated by detection of its mRNA by reverse transcriptase-polymerase chain reaction. The results indicate that *Synechocystis* sp. PCC 6803 **heme oxygenase** encoded by hol can substitute for the defective HY1 gene product and that the only required enzyme activity of the HY1 gene product is **heme oxygenase**.

ST *Synechocystis* gene hol **heme oxygenase** complement phytochrome deficient *Arabidopsis*; cloning *Synechocystis* **heme oxygenase** complement phytochrome deficient *Arabidopsis* hyl

IT Complementation (genetic)
Molecular cloning
 (*Synechocystis* sp. PCC 6803 **heme oxygenase**-encoding hol gene complements phytochrome-deficient *Arabidopsis thaliana* hyl mutant)

IT Phytochromes
 RL: BSU (Biological study, unclassified); BIOL (Biological study)
 (*Synechocystis* sp. PCC 6803 **heme oxygenase**-encoding hol gene complements phytochrome-deficient *Arabidopsis thaliana* hyl mutant)

IT Snapdragon (*Antirrhinum majus*)
 (chimeric oli-hol gene, which contains chloroplast transit peptide-encoding sequence of *Antirrhinum majus* oli gene fused to *Synechocystis* PCC 6803 hol gene, used to transform phytochrome-deficient *Arabidopsis thaliana* hyl mutant)

IT Gene, microbial
 RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
 (hol; *Synechocystis* sp. PCC 6803 **heme oxygenase**-encoding hol gene complements phytochrome-deficient *Arabidopsis thaliana* hyl mutant)

IT Chimeric gene
 RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
 (oli-hol; chimeric oli-hol gene, which contains chloroplast transit peptide-encoding sequence of *Antirrhinum majus* oli gene fused to *Synechocystis* PCC 6803 hol gene, used to transform phytochrome-deficient *Arabidopsis thaliana* hyl mutant)

IT Gene, plant
 RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
 (oli; chimeric oli-hol gene, which contains chloroplast transit

peptide-encoding sequence of *Antirrhinum majus* oli gene fused to
Synechocystis PCC 6803 hol gene, used to transform phytochrome-
deficient *Arabidopsis thaliana* hyl mutant)

IT Protein sequences
(protein sequence comparison of *Arabidopsis thaliana* HY1 protein,
Synechocystis PCC 6803 **heme oxygenase** and human
heme oxygenase 1)

IT *Synechocystis*
(sp. PCC 6803; *Synechocystis* sp. PCC 6803 **heme oxygenase**-encoding hol gene complements phytochrome-deficient *Arabidopsis thaliana* hyl mutant)

IT *Arabidopsis thaliana*
(transformed; *Synechocystis* sp. PCC 6803 **heme oxygenase**-encoding hol gene complements phytochrome-deficient *Arabidopsis thaliana* hyl mutant)

IT **9059-22-7P, Heme oxygenase**
RL: BAC (Biological activity or effector, except adverse); BPN (Biosynthetic preparation); BSU (Biological study, unclassified); BIOL (Biological study); PREP (Preparation)
(*Synechocystis* sp. PCC 6803 **heme oxygenase**-encoding hol gene complements phytochrome-deficient *Arabidopsis thaliana* hyl mutant)

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- (25) Redei, G; Z Vererbungslehre 1962, V93, P164
- (26) Schluchter, W; J Biol Chem 1997, V272, P13562 HCPLUS
- (27) Terry, M; J Biol Chem 1995, V270, P11111 HCPLUS
- (28) Terry, M; J Biol Chem 1996, V271, P21681 HCPLUS
- (29) Terry, M; Plant Cell Environ 1997, V20, P740 HCPLUS
- (30) Weller, J; Plant Cell 1996, V8, P55 HCPLUS
- (31) Weller, J; Plant J 1996, V11, P1177
- (32) Whitelam, G; J Plant Physiol 1991, V139, P119 HCPLUS
- (33) Wilde, A; FEBS Lett 1997, V406, P89 HCPLUS

L79 ANSWER 10 OF 14 HCPLUS COPYRIGHT 2003 ACS
AN 1996:524194 HCPLUS

DN 125:190274

TI The methylotrophic yeast *Pichia pastoris* synthesizes a functionally active chromophore precursor of the plant photoreceptor phytochrome

AU Wu, Shu-Hsing; Lagarias, J. Clark
 CS Section Molecular Cellular Biology, Univ. California, Davis, CA, 95616,
 USA
 SO Proceedings of the National Academy of Sciences of the United States of
 America (1996), 93(17), 8989-8994
 CODEN: PNASA6; ISSN: 0027-8424
 PB National Academy of Sciences
 DT Journal
 LA English
 CC 10-2 (Microbial, Algal, and Fungal Biochemistry)
 Section cross-reference(s): 3
 AB Induction of the expression of an algal phytochrome cDNA in the
 methylotrophic yeast *Pichia pastoris* led to time-dependent formation of
 photoactive **holophytochrome** without the addn. of exogenous
 bilins. Both *in vivo* and *in vitro* difference spectra of this photochromic
 species are very similar to those of higher plant phytochrome A,
 supporting the conclusion that this species possesses a phytochromobilin
 prosthetic group. Zinc blot analyses confirm that a bilin chromophore is
 covalently bound to the algal phytochrome apoprotein. The hypothesis that
P. pastoris contains phytochromobilin synthase, the enzyme that converts
 biliverdin IX. α . to phytochromobilin, was also addressed in this
 study. Sol. exts. from *P. pastoris* were able to convert biliverdin to a
 bilin pigment, which produced a native difference spectrum upon assembly
 with oat apophytochrome A. HPLC analyses confirm that biliverdin is
 converted to both 3E- and 3Z-isomers of phytochromobilin. These
 investigations demonstrate that the ability to synthesize phytochromobilin
 is not restricted to photosynthetic organisms and support the hypothesis
 of a more widespread distribution of the phytochrome photoreceptor.
 ST phytochrome *Pichia*
 IT Molecular cloning
 Pichia pastoris
 (methylotrophic yeast *Pichia pastoris* synthesizes functionally active
 chromophore precursor of plant photoreceptor phytochrome)
 IT Phytochromes
 RL: BSU (Biological study, unclassified); MFM (Metabolic formation); BIOL
 (Biological study); FORM (Formation, nonpreparative)
 (methylotrophic yeast *Pichia pastoris* synthesizes functionally active
 chromophore precursor of plant photoreceptor phytochrome)
 IT 138263-99-7, Phytochromobilin synthase
 RL: BAC (Biological activity or effector, except adverse); BOC (Biological
 occurrence); BSU (Biological study, unclassified); BIOL (Biological
 study); OCCU (Occurrence)
 (*Pichia pastoris* phytochromobilin synthase in relation to phytochrome
 formation)
 IT 114-25-0, Biliverdin
 RL: BPR (Biological process); BSU (Biological study, unclassified); BIOL
 (Biological study); PROC (Process)
 (biliverdin metab. in *Pichia pastoris*)
 IT 78249-71-5, Phytochromobilin
 RL: BSU (Biological study, unclassified); MFM (Metabolic formation); BIOL
 (Biological study); FORM (Formation, nonpreparative)
 (from biliverdin metab. in *Pichia pastoris*)

L79 ANSWER 11 OF 14 HCPLUS COPYRIGHT 2003 ACS
 AN 1995:597221 HCPLUS

DN 123:250826
 TI Candidate genes for the phycoerythrocyanin . α . subunit lyase and
 biochemical analysis of pecE and pecF interposon mutants
 AU Jung, Linda J.; Chan, Crystal F.; **Glazer, Alexander N.**
 CS Department Molecular Cell Biology, University California, Berkeley, CA,
 94720, USA
 SO Journal of Biological Chemistry (1995), 270(21), 12877-84
 CODEN: JBCHA3; ISSN: 0021-9258

PB American Society for Biochemistry and Molecular Biology
 DT Journal
 LA English
 CC 10-1 (Microbial, Algal, and Fungal Biochemistry)
 Section cross-reference(s): 3, 6
 AB The rod substructures of the Anabaena sp. PCC 7120 phycobilisome contain the light harvesting proteins C-phycocyanin and phycoerythrocyanin (PEC). Even at low light intensities, PEC represents no more than 5% of the phycobilisome protein. The .beta. subunits of both proteins carry thioether-linked **phycocyanobilin** (PCB) at .beta.-Cys-82 and .beta.-Cys-155; however, C-phycocyanin has PCB at .alpha.-Cys-84 whereas PEC .alpha. subunit carries **phycobiliviolin** at this position. The Anabaena sp. PCC 7120 pec operon is made up of five genes. PecB and pecA encode the .beta. and .alpha. subunits of PEC, pecC encodes a linker polypeptide assocd. with PEC in the rod substructure, and pecE and pecF are genes of unknown function that show a high degree of homol. to cpcE and cpcF, that encode a C-phycocyanin .alpha. subunit PCB lyase (Fairchild, C. D., Zhao, J., Zhou, J., Colson, S. E., Bryant, D. A., and Glazer, A. N. (1992) Proc. Natl. Acad. Sci. U.S.A. 89, 7017-7021). Insertional mutants in pecE and pecF, and an interposon mutant in which a portion of both pecE and pecF was deleted, were constructed. All three types of mutants grew 1.3 times slower than wild-type under limiting light conditions and showed a 20% redn. in the PCB content of whole cells relative to chlorophyll a. **Holo**-PEC was missing from the phycobilisomes of all three types of mutants and the level of the PEC linker polypeptide was reduced relative to the PEC linker polypeptide was reduced relative to the wild-type. However, .apprx.30% of the wild-type level of the PEC .beta. subunit was present in all of these phycobilisomes. In contrast, the PEC .alpha. subunit was barely detectable in the pecE and pecF mutants, but was present in the pecEF deletion mutant as a PCB-adduct in a 1:1 ratio with the PEC .beta. subunit. The identity of this "unnatural" adduct was confirmed by isolation of the subunit and amino-terminal sequencing. These biochem. results support the inference that pecE and pecF encode a PEC .alpha. subunit **phycobiliviolin** lyase, and, in conjunction with earlier findings, demonstrate that phycobiliprotein bilin lyases show high selectivity (rather than abs. specificity) for both the bilin and the polypeptide substrate.
 ST phycoerythrocyanin alpha subunit lyase pecE pecF; Anabaena phycoerythrocyanin subunit lyase gene pec
 IT Anabaena
 Phycobilisome
 (candidate genes for phycoerythrocyanin .alpha. subunit lyase and biochem. anal. of pecE and pecF interposon mutants)
 IT Gene, microbial
 RL: BSU (Biological study, unclassified); BIOL (Biological study)
 (pecE; candidate genes for phycoerythrocyanin .alpha. subunit lyase and biochem. anal. of pecE and pecF interposon mutants)
 IT Gene, microbial
 RL: BSU (Biological study, unclassified); BIOL (Biological study)
 (pecF; candidate genes for phycoerythrocyanin .alpha. subunit lyase and biochem. anal. of pecE and pecF interposon mutants)
 IT **Biliproteins**
 RL: BSU (Biological study, unclassified); BIOL (Biological study)
 (**phycoerythrocyanins**, candidate genes for phycoerythrocyanin .alpha. subunit lyase and biochem. anal. of pecE and pecF interposon mutants)
 IT 168680-20-4, Phycoerythrocyanin .alpha.-subunit lyase
 RL: BSU (Biological study, unclassified); BIOL (Biological study)
 (candidate genes for phycoerythrocyanin .alpha. subunit lyase and biochem. anal. of pecE and pecF interposon mutants)

AN 1992:628825 HCPLUS
 DN 117:228825
 TI **Phycocyanin .alpha.-subunit**
phycocyanobilin lyase
 AU Fairchild, Craig D.; Zhao, Jindong; Zhou, Jianhui; Colson, Sue Ellen;
 Bryant, Donald A.; **Glazer, Alexander N.**
 CS Dep. Mol. Cell Biol., Univ. California, Berkeley, CA, 94720, USA
 SO Proceedings of the National Academy of Sciences of the United States of
 America (1992), 89(15), 7017-21
 CODEN: PNASA6; ISSN: 0027-8424
 DT Journal
 LA English
 CC 7-2 (Enzymes)
 Section cross-reference(s): 3
 AB Phycobiliproteins, unlike other light-harvesting proteins involved in photosynthesis, bear covalently attached chromophores. The bilin chromophores are attached through thioether bonds to cysteine residues. The cyanobacterium *Synechococcus* sp. PCC7002 has eight distinct bilin attachment sites on seven polypeptides, all of which carry the same chromophore, **phycocyanobilin**. When two genes in the phycocyanin operon of this organism, cpcE and cpcF, are inactivated by insertion, together or sep., the surprising result is elimination of correct bilin attachment at only one site, that on the .alpha. subunit of phycocyanin. CpcE and CpcF were overproduced in *Escherichia coli*. In vitro, these proteins catalyze the attachment of **phycocyanobilin** to the .alpha. subunit of apophycocyanin at the appropriate site, .alpha.-Cys-84, to form the correct adduct. CpcE and CpcF also efficiently catalyze the reverse reaction, in which the bilin from **holo-.alpha.** subunit is transferred either to the apo-.alpha. subunit of the same C-phycocyanin or to the apo-.alpha. subunit of a heterologous C-phycocyanin. The forward and reverse reactions each require both CpcE and CpcF and are specific for the .alpha.-Cys-84 position. **Phycocyanobilin** is the immediate precursor of the protein-bound bilin.
 ST *Synechococcus* phycocyanin subunit **phycocyanobilin** lyase; gene
 cpcE cpcF phycocyanin **phycocyanobilin** lyase
 IT *Synechococcus*
 (**phycocyanin .alpha.-subunit**
 phycocyanobilin lyase of, genes cpcE and cpcF
 encoding and specificity of)
 IT **Phycocyanins**
 RL: BIOL (Biological study)
 (**phycocyanobilin** attachment in, of *Synechococcus*, site of and
 enzyme specific for)
 IT Gene, microbial
 RL: BIOL (Biological study)
 (cpcF, **phycocyanin .alpha.-subunit**
 phycocyanobilin lyase component encoded by, of
 Synechococcus)
 IT Gene, microbial
 RL: BIOL (Biological study)
 (cpcE, **phycocyanin .alpha.-subunit**
 phycocyanobilin lyase component encoded by, of
 Synechococcus)
 IT 144378-42-7, **Phycocyanin .alpha.-subunit**
 phycocyanobilin lyase
 RL: BIOL (Biological study)
 (genes cpcE- and cpcF-encoded, of *Synechococcus*, attachment site
 specificity of)
 IT 52-90-4, Cysteine, biological studies
 RL: BIOL (Biological study)
 (of phycocyanin .alpha. subunit position 84, of *Synechococcus*, as
 phycocyanobilin attachment site)
 IT 20298-86-6, **Phycocyanobilin**

RL: BIOL (Biological study)
 (of phycocyanin, attachment of, by enzyme of Synechococcus,
 localization of binding site in)

L79 ANSWER 13 OF 14 HCPLUS COPYRIGHT 2003 ACS
 AN 1992:210724 HCPLUS
 DN 116:210724
 TI Detection of analytes using fluorescent energy transfer
 IN Tsien, Roger Y.; Taylor, Susan S.; Adams, Stephen R.; Ji, Ying
 PA University of California, Oakland, USA
 SO PCT Int. Appl., 38 pp.
 CODEN: PIXXD2
 DT Patent
 LA English
 IC ICM C12Q001-68
 ICS G01N033-566; G01N033-533
 CC 9-5 (Biochemical Methods)
 Section cross-reference(s): 80

FAN.CNT 1

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	WO 9200388	A1	19920109	WO 1991-US4676	19910701
	W: JP				
	RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LU, NL, SE				
	EP 537270	A1	19930421	EP 1991-913255	19910701
	EP 537270	B1	19980909		
	R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE				
	JP 05508772	T2	19931209	JP 1991-512652	19910701
	JP 3208486	B2	20010910		
	AT 170980	E	19980915	AT 1991-913255	19910701
	US 5439797	A	19950808	US 1993-114103	19930830
PRAI	US 1990-547990	A	19900702		
	WO 1991-US4676	W	19910701		

AB Analytes such as cAMP, GTP, hormone-receptor complexes, Ca²⁺, diacylglycerol, and phorbol esters are detd. by a method involving radiationless energy transfer between 2 fluorochromes, each bound to a protein; the proteins are reversibly assoccd. with one another, the equil. between assocd. and dissocoed. states being dependent on the analyte concn. Thus, the catalytic subunit of cAMP-dependent protein kinase (I) was labeled with FITC and the regulatory subunit of I with tetramethylrhodamine isothiocyanate, and the 2 subunits were allowed to assoc. to form **holoenzyme**. CAMP was detd. in single smooth muscle cells by microinjection of the cells with doubly labeled **holo-I**, illumination of the cells at 490 nm, and measurement of the ratio of emitted light intensity at 500-530 nm (fluorescein emission) and 580 nm (tetramethylrhodamine emission). The ratio rapidly increased after microinjection of isoproterenol (.beta.2-adrenergic agonist which raises cAMP concn.) or forskolin (adenylate cyclase activator) and decreased by propranolol (.beta.2-adrenergic antagonist). Expression of the genes for the catalytic and regulatory subunits of I in Escherichia coli, purifn. of the recombinant subunits, and purifn. of the catalytic subunit of I from porcine heart for use in the cAMP assay are described.

ST fluorescent energy transfer biochem analysis; protein fluorochrome radiationless energy transfer; cAMP detn fluorometry energy transfer

IT Plasmid and Episome
 (62C12, gene for cAMP-dependent protein kinase regulatory subunit on, cloning and expression of, in Escherichia coli)

IT Heart, composition
 (cAMP-dependent protein kinase catalytic subunit of, purifn. of)

IT Fluorescent substances
 (conjugates with proteins, in fluorometric biochem. anal., radiationless energy transfer between fluorochromes in relation to)

IT Gene, animal

IT Receptors
 RL: ANST (Analytical study)
 (for cAMP-dependent protein kinase catalytic and regulatory subunits,
 cloning and expression of, in Escherichia coli)

IT Calmodulins
 RL: ANST (Analytical study)
 (in fluorometric biochem. anal., radiationless energy transfer between
 fluorochromes in relation to)

IT Molecular cloning
 (of cAMP-dependent protein kinase catalytic and regulatory subunit
 genes, in Escherichia coli)

IT Plasmid and Episome
 (pLWS-3, gene for cAMP-dependent protein kinase catalytic subunit on,
 cloning and expression of, in Escherichia coli)

IT Hormones
 RL: ANST (Analytical study)
 (receptors for, detn. of, fluorometric, radiationless energy transfer
 between protein-bound fluorochromes in)

IT Proteins, specific or class
 RL: ANST (Analytical study)
 (GTP-binding, fluorochrome-labeled subunits of, in fluorometric
 biochem. anal., radiationless energy transfer between fluorochromes in
 relation to)

IT Enzymes
 Proteins, specific or class
 RL: ANST (Analytical study)
 (conjugates, with fluorochromes, in fluorometric biochem. anal.,
 radiationless energy transfer between fluorochromes in relation to)

IT Allophycocyanins
 Phycoerythrins
 RL: ANST (Analytical study)
 (conjugates, with proteins, in fluorometric biochem. anal.,
 radiationless energy transfer between fluorochromes in relation to)

IT Glycerides, analysis
 RL: ANT (Analyte); ANST (Analytical study)
 (di-, detn. of, fluorometric, radiationless energy transfer between
 protein-bound fluorochromes in)

IT Spectrochemical analysis
 (fluorometric, fluorescent-labeled proteins in, radiationless energy
 transfer between fluorochromes in relation to)

IT Muscle, composition
 (smooth, cAMP detn. in, fluorescent-labeled cAMP-dependent protein
 kinase catalytic and regulatory subunits in, radiationless energy
 transfer between fluorochromes in relation to)

IT 27072-45-3, FITC
 RL: ANST (Analytical study)
 (cAMP-dependent protein kinase catalytic subunit conjugation with)

IT 107347-53-5, Tetramethylrhodamine isothiocyanate
 RL: ANST (Analytical study)
 (cAMP-dependent protein kinase regulatory subunit conjugation with)

IT 60-92-4, CAMP 86-01-1, GTP 7440-70-2, Calcium, analysis 17673-25-5D,
 Phorbol, esters
 RL: ANT (Analyte); ANST (Analytical study)
 (detn. of, fluorometric, radiationless energy transfer between
 protein-bound fluorochromes in)

IT 2321-07-5D, Fluorescein, protein conjugates 2768-89-0D, Rhodamine X,
 protein conjugates 9026-43-1D, fluorochrome conjugates 19063-57-1D,
 7-Aminocoumarin, derivs., protein conjugates 70281-37-7,
 Tetramethylrhodamine 127409-15-8D, derivs., protein conjugates
 141229-14-3D, protein conjugates

RL: ANST (Analytical study)
 (in fluorometric biochem. anal., radiationless energy transfer between
 fluorochromes in relation to)

IT 142008-29-5P
 RL: PREP (Preparation)
 (purifn. of recombinant catalytic and regulatory subunits of, from
Escherichia coli)

L79 ANSWER 14 OF 14 HCPLUS COPYRIGHT 2003 ACS
 AN 1992:52545 HCPLUS
 DN 116:52545
 TI Expression and assembly of spectrally active recombinant
holophytochrome
 AU Wahleithner, Jill A.; Li, Liming; Lagarias, J. Clark
 CS Dep. Biochem. Biophys., Univ. California, Davis, CA, 95616, USA
 SO Proceedings of the National Academy of Sciences of the United States of
 America (1991), 88(23), 10387-91
 CODEN: PNASA6; ISSN: 0027-8424
 DT Journal
 LA English
 CC 3-2 (Biochemical Genetics)
 Section cross-reference(s): 11
 AB To develop an *in vitro* phytochrome assembly system, the authors expressed
 an oat phytochrome cDNA in both the yeast *Saccharomyces cerevisiae* and the
 bacterium *Escherichia coli*. Anal. of sol. protein exts. showed that the
 recombinant apophytochromes were full-length and capable of covalently
 attaching the phytochrome chromophore analog **phycocyanobilin**.
 Difference spectra indicated that *in vitro*-assembled
holophytochrome species were photoreversible; however, max. and
 min. difference absorption values were blue-shifted relative to those of
 the native photoreceptor. Ext. contg. the recombinant apophytochromes
 were also incubated with phytochromobilin, the natural chromophore
 synthesized from biliverdin by cucumber etioplast prepns. In these
 expts., the difference spectrum obtained was identical to that of native
 oat **holophytochrome**. These results suggest that the recombinant
 apophytochromes adopt a structure similar to that of the apoprotein
 biosynthesized *in vivo*. ELISAs were used to quantitate phytochrome
 expression levels in both yeast and *E. coli* exts. These measurements show
 that 62-75% of the phytochrome apoprotein in the sol. protein ext. was
 competent to assemble with bilins to form spectrally active
holophytochrome.
 ST oat recombinant spectrally active **holophytochrome** assembly;
Saccharomyces cloning recombinant oat phytochrome gene; *Escherichia*
 cloning recombinant oat phytochrome gene; **phycocyanobilin**
 phytochromobilin assembly oat recombinant apophytochrome; phytochrome
holo expression assembly oat yeast

IT Bile pigments
 RL: BIOL (Biological study)
 (assembly of recombinant oat phytochrome apoprotein with, after
 expression in *Escherichia coli* and yeast, for generation of spectrally
 active **holophytochrome**)

IT **Phytochromes**
 RL: BIOL (Biological study)
 (assembly of spectrally active recombinant, of oat, after expression in
Escherichia coli and yeast)

IT *Escherichia coli*
 (cloning and expression in, of apophytochrome phyA3 gene of oat,
 assembly of spectrally active recombinant **holophytochrome**
 subsequent to)

IT *Saccharomyces cerevisiae*
 (cloning and expression in, of apophytochrome phyA3 gene of oats,
 assembly of spectrally active recombinant **holophytochrome**
 subsequent to)

IT Gene, plant
 RL: BIOL (Biological study)
 (for apophytochrome phyA3, of oat, expression in Escherichia coli and yeast of, assembly of spectrally active **holophytochrome** subsequent to)

IT Molecular cloning
 (of oat apophytochrome phyA3 coding region, in Escherichia coli and yeast, assembly of spectrally active **holophytochrome** subsequent to)

IT Plasmid and Episome
 (pGphyA3, oat apophytochrome phyA3 gene on, expression in Escherichia coli of, assembly of spectrally active recombinant **holophytochrome** subsequent to)

IT Plasmid and Episome
 (pMphyA3, oat apophytochrome phyA3 gene on, expression in yeast of, assembly of spectrally active recombinant **holophytochrome** subsequent to)

IT Oat
 (recombinant **holophytochrome** of, assembly of spectrally active, after expression in Escherichia coli and yeast)

IT 20298-86-6, Phycocyanobilin 78249-71-5,
 Phytochromobilin
 RL: BIOL (Biological study)
 (assembly of recombinant oat phytochrome apoprotein with, after expression in Escherichia coli and yeast, for generation of spectrally active **holophytochrome**)

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 E1 THROUGH E8 ASSIGNED

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 DICTIONARY FILE UPDATES: 26 MAR 2003 HIGHEST RN 500755-46-4

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Experimental and calculated property data are now available. See HELP PROPERTIES for more information. See STNote 27, Searching Properties in the CAS Registry File, for complete details:
<http://www.cas.org/ONLINE/STN/STNOTES/stnotes27.pdf>

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 (347401-12-1/RN)
 1 14875-96-8/BI
 (14875-96-8/RN)
 1 20298-86-6/BI

(20298-86-6/RN)
1 168680-20-4/BI
(168680-20-4/RN)
1 124861-40-1/BI
(124861-40-1/RN)
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(144378-42-7/RN)
1 93527-36-7/BI
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-36-7/BI)

=> d ide can tot

L80 ANSWER 1 OF 8 REGISTRY COPYRIGHT 2003 ACS
RN **347401-12-1** REGISTRY
CN Oxidoreductase, ferredoxin:3Z-phycocyanobilin (9CI) (CA INDEX NAME)
OTHER NAMES:
CN 3Z-Phycocyanobilin:ferredoxin oxidoreductase
CN Ferredoxin:3Z-phycocyanobilin oxidoreductase
MF Unspecified
CI MAN
SR CA
LC STN Files: CA, CAPLUS, USPATFULL

*** STRUCTURE DIAGRAM IS NOT AVAILABLE ***
7 REFERENCES IN FILE CA (1962 TO DATE)
7 REFERENCES IN FILE CAPLUS (1962 TO DATE)

REFERENCE 1: 138:181625
REFERENCE 2: 138:148684
REFERENCE 3: 137:365572
REFERENCE 4: 136:180441
REFERENCE 5: 136:50278
REFERENCE 6: 136:2664
REFERENCE 7: 135:73274

L80 ANSWER 2 OF 8 REGISTRY COPYRIGHT 2003 ACS
RN **168680-20-4** REGISTRY
CN Synthase, holophycoerythrocyanin .alpha. subunit (9CI) (CA INDEX NAME)
OTHER NAMES:
CN Phycoerythrobilin lyase
CN Phycoerythrocyanin .alpha.-subunit lyase
CN Phycoerythrocyanin lyase
MF Unspecified
CI MAN
SR CA
LC STN Files: CA, CAPLUS, USPATFULL

*** STRUCTURE DIAGRAM IS NOT AVAILABLE ***
5 REFERENCES IN FILE CA (1962 TO DATE)
6 REFERENCES IN FILE CAPLUS (1962 TO DATE)

REFERENCE 1: 138:181625
REFERENCE 2: 138:148684

REFERENCE 3: 135:340760

REFERENCE 4: 132:331225

REFERENCE 5: 123:250826

L80 ANSWER 3 OF 8 REGISTRY COPYRIGHT 2003 ACS

RN **144378-42-7** REGISTRY

CN Synthase, holophycocyanin .alpha. subunit (9CI) (CA INDEX NAME)

OTHER NAMES:

CN Phycocyanin .alpha.-subunit phycocyanobilin lyase

MF Unspecified

CI MAN

SR CA

LC STN Files: BIOSIS, CA, CAPLUS

*** STRUCTURE DIAGRAM IS NOT AVAILABLE ***

5 REFERENCES IN FILE CA (1962 TO DATE)

5 REFERENCES IN FILE CAPLUS (1962 TO DATE)

REFERENCE 1: 136:2664

REFERENCE 2: 133:330194

REFERENCE 3: 130:234673

REFERENCE 4: 121:3687

REFERENCE 5: 117:228825

L80 ANSWER 4 OF 8 REGISTRY COPYRIGHT 2003 ACS

RN **124861-40-1** REGISTRY

CN Phycobiliviolin (9CI) (CA INDEX NAME)

MF Unspecified

CI MAN

SR CA

LC STN Files: AGRICOLA, BIOSIS, CA, CAPLUS, MEDLINE, USPATFULL

*** STRUCTURE DIAGRAM IS NOT AVAILABLE ***

6 REFERENCES IN FILE CA (1962 TO DATE)

6 REFERENCES IN FILE CAPLUS (1962 TO DATE)

REFERENCE 1: 138:181625

REFERENCE 2: 138:148684

REFERENCE 3: 127:31653

REFERENCE 4: 114:140070

REFERENCE 5: 112:194035

REFERENCE 6: 112:50824

L80 ANSWER 5 OF 8 REGISTRY COPYRIGHT 2003 ACS

RN **93527-36-7** REGISTRY

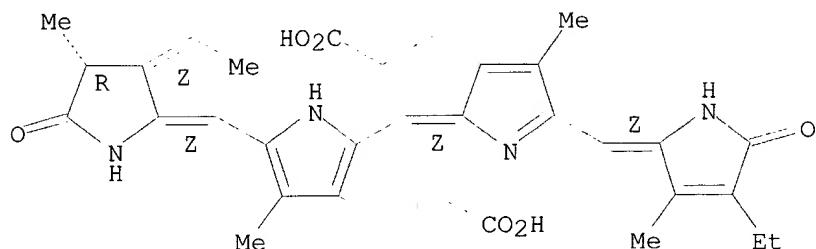
CN 21H-Biline-8,12-dipropanoic acid, 18-ethyl-3-ethylidene-1,2,3,19,22,24-hexahydro-2,7,13,17-tetramethyl-1,19-dioxo-, (2R,3Z)- (9CI) (CA INDEX NAME)

OTHER NAMES:

CN 3(Z)-Phycocyanobilin

FS STEREOSEARCH

MF C33 H38 N4 O6

LC STN Files: AGRICOLA, BEILSTEIN*, BIOSIS, CA, CAPLUS, USPATFULL
(*File contains numerically searchable property data)Absolute stereochemistry.
Double bond geometry as shown.

PROPERTY DATA AVAILABLE IN THE 'PROP' FORMAT

7 REFERENCES IN FILE CA (1962 TO DATE)
7 REFERENCES IN FILE CAPLUS (1962 TO DATE)

REFERENCE 1: 138:148684

REFERENCE 2: 136:199818

REFERENCE 3: 136:2664

REFERENCE 4: 127:343697

REFERENCE 5: 116:37656

REFERENCE 6: 115:275429

REFERENCE 7: 102:2991

L80 ANSWER 6 OF 8 REGISTRY COPYRIGHT 2003 ACS

RN 20298-86-6 REGISTRY

CN 21H-Biline-8,12-dipropionic acid, 18-ethyl-3-ethyldene-1,2,3,19,22,24-hexahydro-2,7,13,17-tetramethyl-1,19-dioxo-, (2R,3E)- (9CI) (CA INDEX NAME)

OTHER CA INDEX NAMES:

CN Biline-8,12-dipropionic acid, 18-ethyl-3-ethyldene-1,2,3,19,22,24-hexahydro-2,7,13,17-tetramethyl-1,19-dioxo- (8CI)

CN Phycocyanobilin (6CI, 7CI)

OTHER NAMES:

CN 3(E)-Phycocyanobilin

AR 20714-57-2

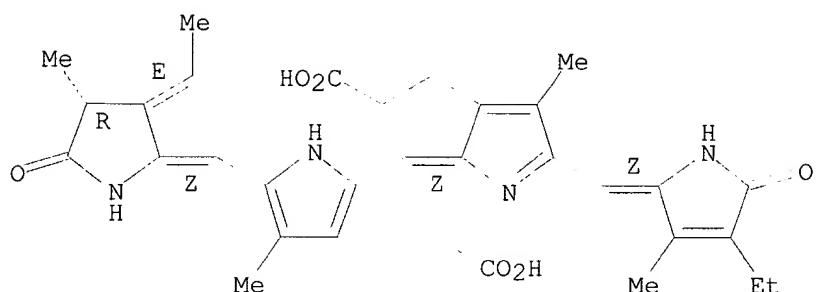
FS STEREOSEARCH

DR 16883-97-9, 18159-29-0, 86747-24-2, 32140-34-4

MF C33 H38 N4 O6

CI COM

LC STN Files: AGRICOLA, BEILSTEIN*, BIOSIS, CA, CANCERLIT, CAOLD, CAPLUS, DDFU, DRUGU, MEDLINE, TOXCENTER
(*File contains numerically searchable property data)Absolute stereochemistry.
Double bond geometry as shown.



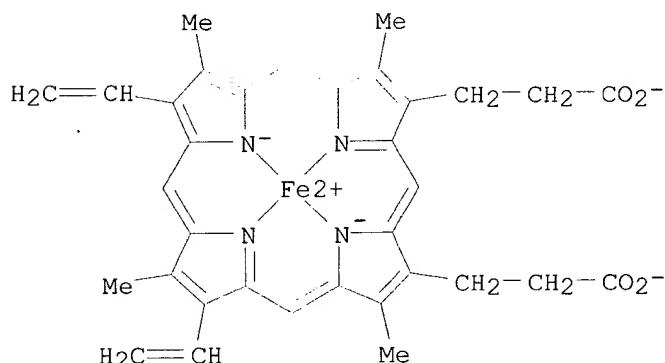
PROPERTY DATA AVAILABLE IN THE 'PROP' FORMAT

179 REFERENCES IN FILE CA (1962 TO DATE)
 13 REFERENCES TO NON-SPECIFIC DERIVATIVES IN FILE CA
 180 REFERENCES IN FILE CAPLUS (1962 TO DATE)
 3 REFERENCES IN FILE CAOLD (PRIOR TO 1967)

REFERENCE 1: 138:181625
 REFERENCE 2: 138:132853
 REFERENCE 3: 138:68600
 REFERENCE 4: 138:21090
 REFERENCE 5: 137:275175
 REFERENCE 6: 137:229409
 REFERENCE 7: 137:151460
 REFERENCE 8: 137:121256
 REFERENCE 9: 137:89372
 REFERENCE 10: 137:44332

L80 ANSWER 7 OF 8 REGISTRY COPYRIGHT 2003 ACS
 RN **14875-96-8** REGISTRY
 CN Ferrate(2-), [7,12-diethenyl-3,8,13,17-tetramethyl-21H,23H-porphine-2,18-dipropanoato(4-)-.kappa.N21,.kappa.N22,.kappa.N23,.kappa.N24]-, dihydrogen, (SP-4-2)- (9CI) (CA INDEX NAME)
 OTHER CA INDEX NAMES:
 CN 21H,23H-Porphine, ferrate(2-) deriv.
 CN 21H,23H-Porphine-2,18-dipropanoic acid, 7,12-diethenyl-3,8,13,17-tetramethyl-, iron complex
 CN Ferrate(2-), [7,12-diethenyl-3,8,13,17-tetramethyl-21H,23H-porphine-2,18-dipropanoato(4-)-N21,N22,N23,N24]-, dihydrogen, (SP-4-2)-
 CN Protoporphyrin, iron complex (6CI)
 OTHER NAMES:
 CN 1,3,5,8-Tetramethyl-2,4-divinylporphine-6,7-dipropionic acid ferrous complex
 CN Ferroheme
 CN Ferroprotoporphyrin IX
 CN Hem Fe
 CN Heme
 CN Heme b
 CN Iron protoporphyrin
 CN Iron protoporphyrin IX
 CN Iron(II) protoporphyrin IX

CN Protoheme
 CN Protoheme IX
 CN Protoporphyrin IX, iron deriv.
 CN Reduced hematin
 CN [Dihydrogen 3,7,12,17-tetramethyl-8,13-divinyl-2,18-porphinedipropionato(2-
)]iron
 DR 86-11-3, 69344-57-6, 75197-11-4
 MF C34 H30 Fe N4 O4 . 2 H
 CI CCS, COM
 LC STN Files: ADISNEWS, AGRICOLA, ANABSTR, BIOBUSINESS, BIOSIS, BIOTECHNO,
 CA, CABAB, CANCERLIT, CAOLD, CAPLUS, CEN, CHEMCATS, CHEMLIST, CIN, DDFU,
 DRUGU, EMBASE, IFICDB, IFIPAT, IFIUDB, IPA, MEDLINE, MRCK*, NIOSHTIC,
 PIRA, PROMT, TOXCENTER, USPATFULL
 (*File contains numerically searchable property data)
 Other Sources: NDSL**, TSCA**
 (**Enter CHEMLIST File for up-to-date regulatory information)
 CRN (104414-01-9)



● 2 H⁺

8136 REFERENCES IN FILE CA (1962 TO DATE)
 364 REFERENCES TO NON-SPECIFIC DERIVATIVES IN FILE CA
 8164 REFERENCES IN FILE CAPLUS (1962 TO DATE)
 3 REFERENCES IN FILE CAOLD (PRIOR TO 1967)

REFERENCE 1: 138:184613

REFERENCE 2: 138:184252

REFERENCE 3: 138:183658

REFERENCE 4: 138:183001

REFERENCE 5: 138:182997

REFERENCE 6: 138:182996

REFERENCE 7: 138:182995

REFERENCE 8: 138:182991

REFERENCE 9: 138:182973

REFERENCE 10: 138:182788

L80 ANSWER 8 OF 8 REGISTRY COPYRIGHT 2003 ACS
RN 9059-22-7 REGISTRY
CN Oxygenase, heme (decyclizing) (9CI) (CA INDEX NAME)
OTHER NAMES:
CN E.C. 1.14.99.3
CN Heme oxygenase
CN ORP33 proteins
CN Proteins, ORP33 (oxygen-regulated protein, 33,000-mol.-wt.)
MF Unspecified
CI MAN
LC STN Files: ADISNEWS, AGRICOLA, ANABSTR, BIOBUSINESS, BIOSIS, BIOTECHNO,
CA, CAPLUS, CEN, CIN, EMBASE, PROMT, TOXCENTER, USPATFULL

*** STRUCTURE DIAGRAM IS NOT AVAILABLE ***
2206 REFERENCES IN FILE CA (1962 TO DATE)
23 REFERENCES TO NON-SPECIFIC DERIVATIVES IN FILE CA
2214 REFERENCES IN FILE CAPLUS (1962 TO DATE)

REFERENCE 1: 138:186231
REFERENCE 2: 138:185239
REFERENCE 3: 138:185006
REFERENCE 4: 138:184957
REFERENCE 5: 138:183158
REFERENCE 6: 138:182973
REFERENCE 7: 138:181625
REFERENCE 8: 138:180708
REFERENCE 9: 138:180623
REFERENCE 10: 138:180555

=> d ide can tot 181

L81 ANSWER 1 OF 10 REGISTRY COPYRIGHT 2003 ACS
RN 347401-21-2 REGISTRY
CN Oxidoreductase, ferredoxin:3Z-phycoerythrobilin (9CI) (CA INDEX NAME)
OTHER NAMES:
CN Ferredoxin:3Z-phycoerythrobilin oxidoreductase
MF Unspecified
CI MAN
SR CA
LC STN Files: CA, CAPLUS, USPATFULL

*** STRUCTURE DIAGRAM IS NOT AVAILABLE ***
4 REFERENCES IN FILE CA (1962 TO DATE)
4 REFERENCES IN FILE CAPLUS (1962 TO DATE)

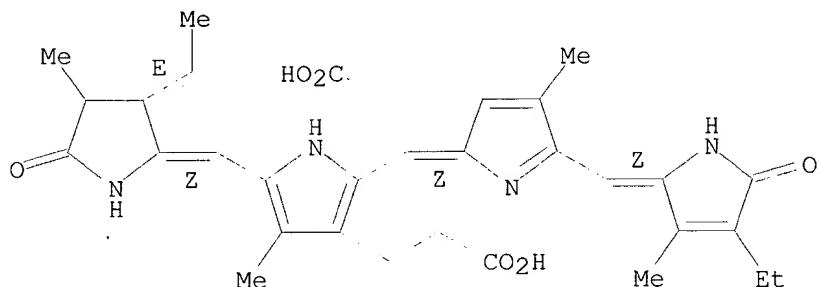
REFERENCE 1: 138:148684
REFERENCE 2: 137:365572
REFERENCE 3: 136:50278
REFERENCE 4: 135:73274

L81 ANSWER 2 OF 10 REGISTRY COPYRIGHT 2003 ACS
 RN 215871-76-4 REGISTRY
 CN 21H-Biline-8,12-dipropanoic acid, 18-ethyl-3-ethyldene-
 1,2,3,19,22,24-hexahydro-2,7,13,17-tetramethyl-1,19-dioxo-, (3E)- (9CI)
 (CA INDEX NAME)

OTHER NAMES:

CN (.+-.)-Phycocyanobilin
 FS STEREOSEARCH
 DR 322642-34-2
 MF C33 H38 N4 O6
 SR CA
 LC STN Files: CA, CAPLUS, CASREACT

Double bond geometry as shown.



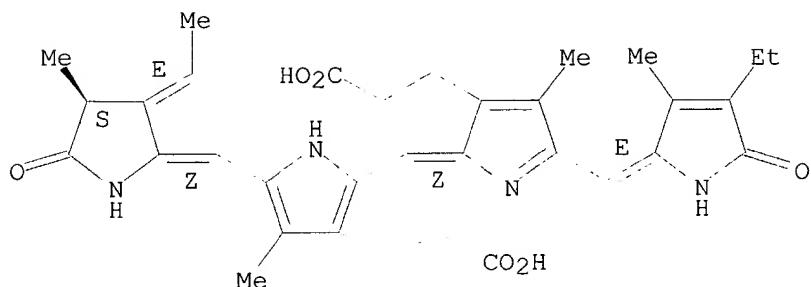
PROPERTY DATA AVAILABLE IN THE 'PROP' FORMAT

7 REFERENCES IN FILE CA (1962 TO DATE)
 7 REFERENCES IN FILE CAPLUS (1962 TO DATE)

REFERENCE 1: 135:1857
 REFERENCE 2: 134:147428
 REFERENCE 3: 133:120180
 REFERENCE 4: 132:279038
 REFERENCE 5: 130:153494
 REFERENCE 6: 130:3704

L81 ANSWER 3 OF 10 REGISTRY COPYRIGHT 2003 ACS
 RN 214054-28-1 REGISTRY
 CN 21H-Biline-8,12-dipropanoic acid, 18-ethyl-3-ethyldene-
 1,2,3,19,22,24-hexahydro-2,7,13,17-tetramethyl-1,19-dioxo-, (2S,3E,15E)-
 (9CI) (CA INDEX NAME)
 FS STEREOSEARCH
 MF C33 H38 N4 O6
 SR CA
 LC STN Files: CA, CAPLUS

Absolute stereochemistry.
 Double bond geometry as shown.



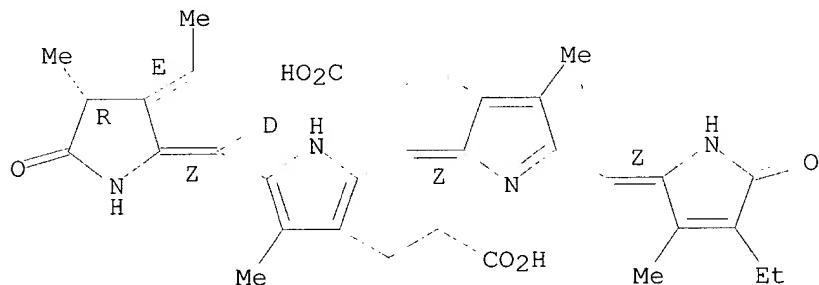
PROPERTY DATA AVAILABLE IN THE 'PROP' FORMAT

1 REFERENCES IN FILE CA (1962 TO DATE)
1 REFERENCES IN FILE CAPLUS (1962 TO DATE)

REFERENCE 1: 129:289975

L81 ANSWER 4 OF 10 REGISTRY COPYRIGHT 2003 ACS
RN 189246-94-4 REGISTRY
CN 21H-Biline-5-d-8,12-dipropionic acid, 18-ethyl-3-ethyldene-
1,2,3,19,22,24-hexahydro-2,7,13,17-tetramethyl-1,19-dioxo-, (2R,3E)-
(9CI) (CA INDEX NAME)
FS STEREOSEARCH
MF C33 H37 D N4 O6
SR CA
LC STN Files: CA, CAPLUS

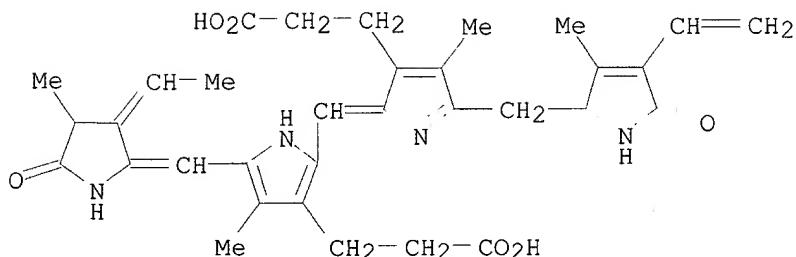
Absolute stereochemistry.
Double bond geometry as shown.



1 REFERENCES IN FILE CA (1962 TO DATE)
1 REFERENCES IN FILE CAPLUS (1962 TO DATE)

REFERENCE 1: 126:317272

L81 ANSWER 5 OF 10 REGISTRY COPYRIGHT 2003 ACS
RN 137332-18-4 REGISTRY
CN 21H-Biline-8,12-dipropionic acid, 18-ethenyl-3-ethyldene-
1,2,3,15,16,19,22,24-octahydro-2,7,13,17-tetramethyl-1,19-dioxo-,
(2R,3Z,16R)- (9CI) (CA INDEX NAME)
MF C33 H38 N4 O6
SR CA
LC STN Files: BEILSTEIN*, CA, CAPLUS
(*File contains numerically searchable property data)



PROPERTY DATA AVAILABLE IN THE 'PROP' FORMAT

2 REFERENCES IN FILE CA (1962 TO DATE)
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REFERENCE 1: 116:37656

REFERENCE 2: 115:275429

L81 ANSWER 6 OF 10 REGISTRY COPYRIGHT 2003 ACS

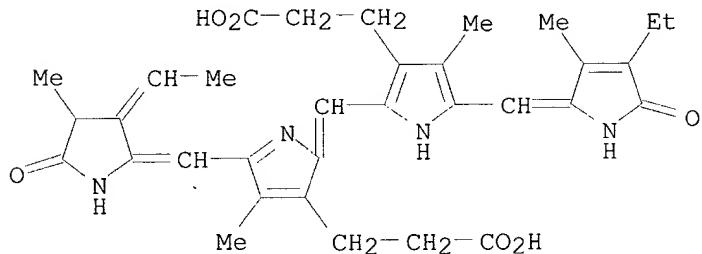
RN 133561-60-1 REGISTRY

CN 21H-Biline-8,12-dipropanoic acid, 18-ethyl-3-ethyldene-1,2,3,19,23,24-hexahydro-2,7,13,17-tetramethyl-1,19-dioxo- (9CI) (CA INDEX NAME)

MF C33 H38 N4 O6

SR CA

LC STN Files: BEILSTEIN*, CA, CAPLUS
 (*File contains numerically searchable property data)



PROPERTY DATA AVAILABLE IN THE 'PROP' FORMAT

1 REFERENCES IN FILE CA (1962 TO DATE)
 1 REFERENCES IN FILE CAPLUS (1962 TO DATE)

REFERENCE 1: 114:201852

L81 ANSWER 7 OF 10 REGISTRY COPYRIGHT 2003 ACS

RN 111565-55-0 REGISTRY

CN Protein LRC27, PC (Nostoc muscorum clone pAn410 27-kilodalton rod-core-linking reduced) (9CI) (CA INDEX NAME)

OTHER NAMES:

CN Phycocyanobilin lyase alpha subunit (Nostoc sp. PCC 7120 gene cpcE)

CN Protein (Anabaena PCC7120 strain PCC7120 gene cpcE)

FS PROTEIN SEQUENCE

MF Unspecified

CI MAN

SR CA
 LC STN Files: CA, CAPLUS

RELATED SEQUENCES AVAILABLE WITH SEQLINK

*** STRUCTURE DIAGRAM IS NOT AVAILABLE ***
 *** USE 'SQD' OR 'SQIDE' FORMATS TO DISPLAY SEQUENCE ***
 3 REFERENCES IN FILE CA (1962 TO DATE)
 3 REFERENCES IN FILE CAPLUS (1962 TO DATE)

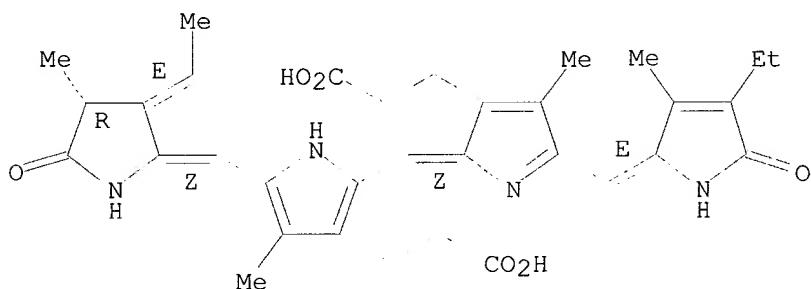
REFERENCE 1: 136:65028

REFERENCE 2: 135:299437

REFERENCE 3: 107:230204

L81 ANSWER 8 OF 10 REGISTRY COPYRIGHT 2003 ACS
 RN 86746-89-6 REGISTRY
 CN 21H-Biline-8,12-dipropanoic acid, 18-ethyl-3-ethyldene-
 1,2,3,19,22,24-hexahydro-2,7,13,17-tetramethyl-1,19-dioxo-, (2R,3E,15E)-
 (9CI) (CA INDEX NAME)
 FS STEREOSEARCH
 MF C33 H38 N4 O6
 LC STN Files: BEILSTEIN*, CA, CAPLUS
 (*File contains numerically searchable property data)

Absolute stereochemistry.
 Double bond geometry as shown.

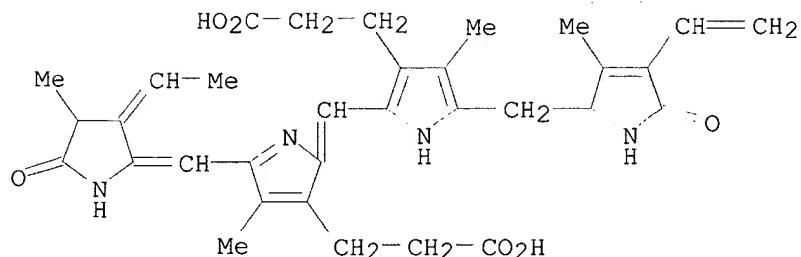


PROPERTY DATA AVAILABLE IN THE 'PROP' FORMAT

1 REFERENCES IN FILE CA (1962 TO DATE)
 1 REFERENCES IN FILE CAPLUS (1962 TO DATE)

REFERENCE 1: 99:83931

L81 ANSWER 9 OF 10 REGISTRY COPYRIGHT 2003 ACS
 RN 71189-94-1 REGISTRY
 CN 21H-Biline-8,12-dipropanoic acid, 18-ethenyl-3-ethyldene-
 1,2,3,15,16,19,23,24-octahydro-2,7,13,17-tetramethyl-1,19-dioxo-,
 (2R,16R)- (9CI) (CA INDEX NAME)
 MF C33 H38 N4 O6
 LC STN Files: CA, CAPLUS



PROPERTY DATA AVAILABLE IN THE 'PROP' FORMAT

1 REFERENCES IN FILE CA (1962 TO DATE)
 1 REFERENCES IN FILE CAPLUS (1962 TO DATE)

REFERENCE 1: 91:107855

L81 ANSWER 10 OF 10 REGISTRY COPYRIGHT 2003 ACS

RN 18097-67-1 REGISTRY

CN 21H-Biline-8,12-dipropionic acid, 18-ethenyl-3-ethylidene-1,2,3,15,16,19,22,24-octahydro-2,7,13,17-tetramethyl-1,19-dioxo-, (2R,3E,16R)- (9CI) (CA INDEX NAME)

OTHER CA INDEX NAMES:

CN Biline-8,12-dipropionic acid, 3-ethylidene-1,2,3,15,16,19,22,24-octahydro-2,7,13,17-tetramethyl-1,19-dioxo-18-vinyl- (8CI)

OTHER NAMES:

CN 3(E)-Phycoerythrobilin

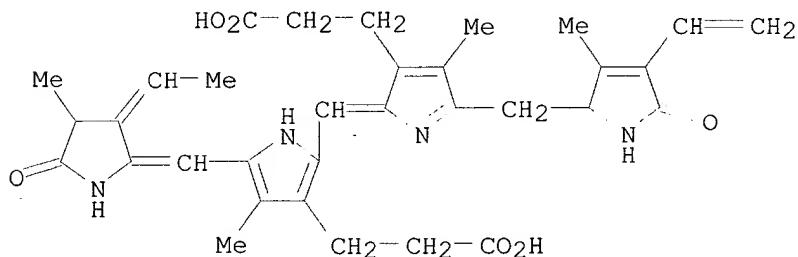
CN Phycoerythrobilin

DR 18159-28-9

MF C33 H38 N4 O6

LC STN Files: AGRICOLA, BEILSTEIN*, BIOSIS, CA, CAPLUS, MEDLINE, TOXCENTER, USPATFULL

(*File contains numerically searchable property data)



PROPERTY DATA AVAILABLE IN THE 'PROP' FORMAT

117 REFERENCES IN FILE CA (1962 TO DATE)
 6 REFERENCES TO NON-SPECIFIC DERIVATIVES IN FILE CA
 117 REFERENCES IN FILE CAPLUS (1962 TO DATE)

REFERENCE 1: 138:148684

REFERENCE 2: 137:301970

REFERENCE 3: 137:275175

REFERENCE 4: 137:151460

REFERENCE 5: 137:89372

REFERENCE 6: 136:243489

REFERENCE 7: 135:354347

REFERENCE 8: 135:340760

REFERENCE 9: 135:270216

REFERENCE 10: 135:73274

=> fil hcaplus

FILE 'HCAPLUS' ENTERED AT 13:55:52 ON 27 MAR 2003
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FILE COVERS 1907 - 27 Mar 2003 VOL 138 ISS 13
FILE LAST UPDATED: 26 Mar 2003 (20030326/ED)

This file contains .CAS Registry Numbers for easy and accurate substance identification.

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L92 ANSWER 1 OF 5 HCAPLUS COPYRIGHT 2003 ACS

AN 2001:597094 HCAPLUS

DN 135:207297

TI Purification and biochemical properties of phytochromobilin synthase from etiolated oat seedlings

AU McDowell, Michael T.; Lagarias, J. Clark

CS Section of Molecular and Cellular Biology, University of California, Davis, CA, 95616, USA

SO Plant Physiology (2001), 126(4), 1546-1554

CODEN: PLPHAY; ISSN: 0032-0889

PB American Society of Plant Physiologists

DT Journal

LA English

CC 7-2 (Enzymes)

AB Plant phytochromes are dependent on the covalent attachment of the linear tetrapyrrole chromophore phytochromobilin (P.PHI.B) for photoactivity. In plants, biliverdin IX.alpha. (BV) is reduced by plastid-localized, ferredoxin (Fd)-dependent phytochromobilin synthase (I) to yield 3Z-P.PHI.B. Here, the >50,000-fold purifn. of I from etioplasts of dark-grown oat (*Avena sativa* L. cv Garry) seedlings is described, using traditional column chromatog. and preparative electrophoresis. Thus, I is a very low abundance enzyme with a robust turnover rate. The turnover rate was estd. to be >100 s⁻¹, which is similar to that of mammalian

NAD(P)H-dependent BV reductase. Oat I was a monomer with a subunit mol. wt. of 29 kDa. However, 2 distinct charged forms of I were identified by native isoelec. focusing. The ability of I to reduce BV was dependent on reduced 2Fe-2S Fd. The Km for spinach Fd was detd. to be 3-4 .mu.M. I had a high affinity for its bilin substrate, with a submicromolar Km value for BV.

ST phytochromobilin synthase oat seedling

IT Michaelis constant

(of phytochromobilin synthase from etiolated oat seedlings)

IT Oat

(purifn. and characterization of phytochromobilin synthase from etiolated oat seedlings)

IT 138263-99-7P, Phytochromobilin synthase

RL: BAC (Biological activity or effector, except adverse); BSU (Biological study, unclassified); PRP (Properties); PUR (Purification or recovery); BIOL (Biological study); PREP (Preparation)
(purifn. and characterization of phytochromobilin synthase from etiolated oat seedlings)

RE.CNT 35 THERE ARE 35 CITED REFERENCES AVAILABLE FOR THIS RECORD

RE

- (1) Ausubel, F; Current Protocols in Molecular Biology 1991
- (2) Beale, S; Chem Rev 1993, V93, P785 HCPLUS
- (3) Beale, S; J Biol Chem 1991, V266, P22328 HCPLUS
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- (7) Blum, H; Electrophoresis 1987, V8, P93 HCPLUS
- (8) Bradford, M; Anal Biochem 1976, V72, P248 HCPLUS
- (9) Buchanan, B; Methods Enzymol 1971, V23, P413 HCPLUS
- (10) Cornejo, J; Photosynth Res 1997, V51, P223 HCPLUS
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- (13) Davis, S; Science 1999, V286, P2517 HCPLUS
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- (15) Hershey, H; Nucleic Acids Res 1985, V13, P8543 HCPLUS
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- (32) Wu, S; J Biol Chem 1997, V272, P25700 HCPLUS
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- (34) Yeh, K; Science 1997, V277, P1505 HCPLUS
- (35) Yoshida, T; J Biol Chem 1979, V254, P4487 HCPLUS

L92 ANSWER 2 OF 5 HCPLUS COPYRIGHT 2003 ACS

AN 2001:378669 HCPLUS

DN 135:118588

TI The Arabidopsis HY2 gene encodes phytochromobilin synthase, a ferredoxin-dependent biliverdin reductase

AU Kohchi, Takayuki; Mukougawa, Keiko; Frankenberg, Nicole; Masuda, Munehisa; Yokota, Akiho; Lagarias, J. Clark

CS Graduate School of Biological Sciences, Nara Institute of Science and
 Technology, Nara, 630-0101, Japan
 SO Plant Cell (2001), 13(2), 425-436
 CODEN: PLCEEW; ISSN: 1040-4651
 PB American Society of Plant Physiologists
 DT Journal
 LA English
 CC 7-2 (Enzymes)
 Section cross-reference(s): 3, 11
 AB Light perception by the plant photoreceptor phytochrome requires the tetrapyrrole chromophore phytochromobilin (P.PHI.B), which is covalently attached to a large apoprotein. *Arabidopsis* mutants *hyl* and *hy2*, which are defective in P.PHI.B biosynthesis, display altered responses to light due to a deficiency in photoactive phytochrome. Here, the authors describe the isolation of the *HY2* gene by map-based cloning. *Hy2* mutant alleles possess alterations within this locus, some of which affect the expression of the *HY2* transcript. *HY2* encodes a sol. protein precursor of 38 kDa with a putative N-terminal plastid transit peptide. The *HY2* transit peptide is sufficient to localize the reporter green fluorescent protein to plastids. Purified mature recombinant *HY2* protein exhibits P.PHI.B synthase activity (i.e., ferredoxin-dependent redn. of biliverdin IX.alpha. to P.PHI.B), as confirmed by HPLC and by the ability of the **bilin** reaction products to combine with apophytochrome to yield photoactive holophytochrome. Database searches and hybridization studies suggest that *HY2* is a unique gene in the *Arabidopsis* genome that is related to a family of proteins found in oxygenic photosynthetic bacteria.
 ST cDNA sequence gene *HY2* phytochromobilin synthase *Arabidopsis*
 IT Gene, plant
 RL: BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study)
 (HY2; cDNA and amino acid sequences of gene *HY2* phytochromobilin synthase of *Arabidopsis thaliana*)
 IT Protein sequences
 cDNA sequences
 (cDNA and amino acid sequences of gene *HY2* phytochromobilin synthase of *Arabidopsis thaliana*)
 IT *Arabidopsis thaliana*
 (gene *HY2* phytochromobilin synthase of *Arabidopsis thaliana* and mol. characterization of gene *HY2*)
 IT Plastid
 (plastid localization of gene *HY2* phytochromobilin synthase of *Arabidopsis thaliana*)
 IT 350869-22-6
 RL: BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study)
 (amino acid sequence; cDNA and amino acid sequences of gene *HY2* phytochromobilin synthase of *Arabidopsis thaliana*)
 IT 138263-99-7, Phytochromobilin synthase
 RL: BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study)
 (gene *HY2*; gene *HY2* phytochromobilin synthase of *Arabidopsis thaliana* and mol. characterization of gene *HY2*)
 IT 328223-94-5, GenBank AB045112
 RL: BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study)
 (nucleotide sequence; cDNA and amino acid sequences of gene *HY2* phytochromobilin synthase of *Arabidopsis thaliana*)
 RE.CNT 52 THERE ARE 52 CITED REFERENCES AVAILABLE FOR THIS RECORD
 RE
 (1) Altschul, S; J Mol Biol 1990, V215, P403 HCAPLUS
 (2) Altschul, S; Nucleic Acids Res 1997, V25, P3389 HCAPLUS
 (3) Beale, S; Arch Biochem Biophys 1984, V235, P371 HCAPLUS
 (4) Beale, S; Chem Rev 1993, V93, P785 HCAPLUS

- (5) Becker, T; *Planta* 1992, V188, P39 HCAPLUS
 (6) Bowler, C; *Cell* 1994, V77, P73 HCAPLUS
 (7) Bowler, C; *Plant Cell* 1994, V6, P1529 HCAPLUS
 (8) Bradford, M; *Anal Biochem* 1976, V72, P248 HCAPLUS
 (9) Brown, J; *Plant J* 1996, V10, P771 HCAPLUS
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L92 ANSWER 3 OF 5 HCAPLUS COPYRIGHT 2003 ACS
 AN 2001:312008 HCAPLUS
 DN 135:73274
 TI Functional genomic analysis of the HY2 family of ferredoxin-dependent bilin reductases from oxygenic photosynthetic organisms
 AU Frankenberg, Nicole; Mukougawa, Keiko; Kohchi, Takayuki; Lagarias, J. Clark
 CS Section of Molecular and Cellular Biology, University of California at Davis, Davis, CA, 95616, USA
 SO Plant Cell (2001), 13(4), 965-978
 CODEN: PLCEEW; ISSN: 1040-4651
 PB American Society of Plant Physiologists
 DT Journal
 LA English

- CC 7-5 (Enzymes)
 Section cross-reference(s): 10, 11
- AB Phytobilins are linear tetrapyrrole precursors of the light-harvesting prosthetic groups of the phytochrome photoreceptors of plants and the **phycobiliprotein** photosynthetic antennae of cyanobacteria, red algae, and cryptomonads. Previous biochem. studies have established that phytobilins are synthesized from **heme** via the intermediacy of biliverdin IX.alpha. (BV), which is reduced subsequently by ferredoxin-dependent **bilin** reductases with different double-bond specificities. By exploiting the sequence of phytochromobilin synthase (HY2) of Arabidopsis, an enzyme that catalyzes the ferredoxin-dependent conversion of BV to the phytochrome chromophore precursor phytochromobilin, genes encoding putative **bilin** reductases were identified in the genomes of various cyanobacteria, oxyphotobacteria, and plants. Phylogenetic analyses resolved four classes of HY2-related genes, one of which encodes red chlorophyll catabolite reductases, which are **bilin** reductases involved in chlorophyll catabolism in plants. To test the catalytic activities of these putative enzymes, representative HY2-related genes from each class were amplified by the polymerase chain reaction and expressed in Escherichia coli. Using a coupled apophytochrome assembly assay and HPLC anal., we exmd. the ability of the recombinant proteins to catalyze the ferredoxin-dependent redn. of BV to phytobilins. These investigations defined three new classes of **bilin** reductases with distinct substrate/product specificities that are involved in the biosynthesis of the **phycobiliprotein** chromophore precursors **phycoerythrobilin** and **phycocyanobilin**. Implications of these results are discussed with regard to the pathways of phytobilin biosynthesis and their evolution.
- ST **bilin** reductase sequence cyanobacteria plant oxyphotobacteria
- IT Cyanobacteria
 Enzyme functional sites
 Oxyphotobacteria
 Plant (Embryophyta)
 Protein sequences
 (functional genomic anal. of HY2 family of ferredoxin-dependent **bilin** reductases from oxygenic photosynthetic organisms)
- IT Evolution
 (mol.; functional genomic anal. of HY2 family of ferredoxin-dependent **bilin** reductases from oxygenic photosynthetic organisms)
- IT Bile pigments
 RL: BPR (Biological process); BSU (Biological study, unclassified); BIOL (Biological study); PROC (Process)
 (phytobilins; functional genomic anal. of HY2 family of ferredoxin-dependent **bilin** reductases from oxygenic photosynthetic organisms)
- IT 138263-99-7, Ferredoxin:3Z-phytochromobilin oxidoreductase
 347401-12-1, Ferredoxin:3Z-phycocyanobilin .
 oxidoreductase 347401-20-1 347401-21-2
 RL: BAC (Biological activity or effector, except adverse); BOC (Biological occurrence); BPR (Biological process); BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study); OCCU (Occurrence); PROC (Process)
 (functional genomic anal. of HY2 family of **ferredoxin**-dependent **bilin** reductases from oxygenic photosynthetic organisms)
- IT 114-25-0, Biliverdin 18097-67-1,
Phycoerythrobilin 20298-86-6, **Phycocyanobilin**
 RL: BPR (Biological process); BSU (Biological study, unclassified); BIOL (Biological study); PROC (Process)
 (functional genomic anal. of HY2 family of ferredoxin-dependent **bilin** reductases from oxygenic photosynthetic organisms)
- RE.CNT 50 THERE ARE 50 CITED REFERENCES AVAILABLE FOR THIS RECORD
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L92 ANSWER 4 OF 5 HCPLUS COPYRIGHT 2003 ACS
 AN 2000:175309 HCPLUS
 DN 132:204723
 TI Purification and characterization of phytochromobilin synthase from Avena sativa
 AU McDowell, Michael Thomas
 CS Univ. of California, Davis, CA, USA
 SO (1999) 147 pp. Avail.: UMI, Order No. DA9940114
 From: Diss. Abstr. Int., B 2000, 60(8), 3931
 DT Dissertation
 LA English

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NEWS 26 Oct 01 CASREACT Enriched with Reactions from 1907 to 1985
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NEWS 28 Oct 24 BEILSTEIN adds new search fields
NEWS 29 Oct 24 Nutraceuticals International (NUTRACEUT) now available on STN
NEWS 30 Oct 25 MEDLINE SDI run of October 8, 2002
NEWS 31 Nov 18 DKILIT has been renamed APOLLIT
NEWS 32 Nov 25 More calculated properties added to REGISTRY
NEWS 33 Dec 02 TIBKAT will be removed from STN

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NEWS 35 Dec 17 PCTFULL now covers WP/PCT Applications from 1978 to date
NEWS 36 Dec 17 TOXCENTER enhanced with additional content
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NEWS 38 Dec 30 ISMEC no longer available
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NEWS 40 Jan 21 PHARMAML offering one free connect hour in February 2003
NEWS 41 Jan 29 Simultaneous left and right truncation added to COMPENDEX,
ENERGY, INSPEC
NEWS 42 Feb 13 CANCERLIT is no longer being updated
NEWS 43 Feb 24 METADEX enhancements
NEWS 44 Feb 24 PCTGEN now available on STN
NEWS 45 Feb 24 TEMA now available on STN

NEWS 46 Feb 26 NTIS now allows simultaneous left and right truncation
NEWS 47 Feb 26 PCTFULL now contains images
NEWS 48 Mar 04 SDI PACKAGE for monthly delivery of multifile SDI results
NEWS 49 Mar 19 APOLLIT offering free connect time in April 2003
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NEWS 52 Mar 24 Additional information for trade-named substances without structures available in REGISTRY
NEWS 53 Mar 24 Indexing from 1957 to 1966 added to records in CA/CAPLUS

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AND CURRENT DISCOVER FILE IS DATED 01 OCTOBER 2002

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=> s 11 and holo-alpha subun

L2 5 L1 AND HOLO-ALPHA SUBUNIT

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L2 ANSWER 1 OF 5 MEDLINE

TI Biosynthesis of a fluorescent cyanobacterial C-
phycocyanin holo-alpha subunit in a
heterologous host.

AB The entire pathway for the synthesis of a fluorescent holophycobiliprotein subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803) was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to the natural chromophore 3Z-phycocyanobilin, namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase, were expressed from a plasmid under control of the hybrid trp-lac (trc) promoter. Genes for the apoprotein (**C-phycocyanin alpha subunit**; cpcA) and the heterodimeric lyase (cpcE and cpcF) that catalyzes chromophore attachment were expressed from the trc promoter on a second plasmid. Upon induction, recombinant *E. coli* used the cellular pool of heme to produce holo-CpcA with spectroscopic properties qualitatively and quantitatively similar to those of the same protein produced endogenously in cyanobacteria. About a third of the apo-CpcA was converted to holo-CpcA. No significant bilin addition took place in a similarly engineered *E. coli* strain that lacks cpcE and cpcF. This approach should permit incisive analysis of many remaining questions in phycobiliprotein biosynthesis. These studies also demonstrate the feasibility of generating constructs of these proteins *in situ* for use as fluorescent protein probes in living cells.

ACCESSION NUMBER: 2001504133 MEDLINE

DOCUMENT NUMBER: 21438034 PubMed ID: 11553806

TITLE: Biosynthesis of a fluorescent cyanobacterial C-
phycocyanin holo-alpha
subunit in a heterologous host.

AUTHOR: Tooley A J; Cai Y A; Glazer A N

CORPORATE SOURCE: Department of Molecular and Cell Biology, University of California, 142 LSA no. 3200, Berkeley, CA 94720-3200, USA.

SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (2001 Sep 11) 98 (19) 10560-5.
Journal code: 7505876. ISSN: 0027-8424.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

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FILE SEGMENT: Priority Journals

ENTRY MONTH: 200111

ENTRY DATE: Entered STN: 20010913

Last Updated on STN: 20011105

Entered Medline: 20011101

L2 ANSWER 2 OF 5 MEDLINE

TI Phycocyanin alpha-subunit phycocyanobilin lyase.

AB Phycobiliproteins, unlike other light-harvesting proteins involved in photosynthesis, bear covalently attached chromophores. The bilin chromophores are attached through thioether bonds to cysteine residues. The cyanobacterium *Synechococcus* sp. PCC 7002 has eight distinct bilin attachment sites on seven polypeptides, all of which carry the same chromophore, phycocyanobilin. When two genes in the phycocyanin operon of this organism, cpcE and cpcF, are inactivated by insertion, together or separately, the surprising result is elimination of correct bilin attachment at only one site, that on the alpha subunit of phycocyanin. We have overproduced CpcE and CpcF in *Escherichia coli*. In vitro, these proteins catalyze the attachment of phycocyanobilin to the alpha subunit of apophycocyanin at the appropriate site, alpha-Cys-84, to form the correct adduct. CpcE and CpcF also efficiently catalyze the reverse reaction, in which the bilin from **holo-alpha** subunit is transferred either to the apo-alpha subunit of the same

C-phycocyanin or to the apo-alpha subunit of a heterologous **C-phycocyanin**. The forward and reverse reactions each require both CpcE and CpcF and are specific for the alpha-Cys-84 position. Phycocyanobilin is the immediate precursor of the protein-bound bilin.

ACCESSION NUMBER: 92357762 MEDLINE
DOCUMENT NUMBER: 92357762 PubMed ID: 1495995
TITLE: Phycocyanin alpha-subunit phycocyanobilin lyase.
AUTHOR: Fairchild C D; Zhao J; Zhou J; Colson S E; Bryant D A;
Glazer A N
CORPORATE SOURCE: Department of Molecular and Cell Biology, University of California, Berkeley 94720.
CONTRACT NUMBER: GM28994 (NIGMS)
GM31625 (NIGMS)
SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (1992 Aug 1) 89 (15) 7017-21.
Journal code: 7505876. ISSN: 0027-8424.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199209
ENTRY DATE: Entered STN: 19920925
Last Updated on STN: 19970203
Entered Medline: 19920904

L2 ANSWER 3 OF 5 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
TI Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-alpha subunit in a heterologous host.
AB The entire pathway for the synthesis of a fluorescent holophycobiliprotein subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803) was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to the natural chromophore 3Z-phycocyanobilin, namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase, were expressed from a plasmid under control of the hybrid trp-lac (trc) promoter. Genes for the apoprotein (C-phycocyanin alpha subunit; cpcA) and the heterodimeric lyase (cpcE and cpcF) that catalyzes chromophore attachment were expressed from the trc promoter on a second plasmid. Upon induction, recombinant *E. coli* used the cellular pool of heme to produce holo-CpcA with spectroscopic properties qualitatively and quantitatively similar to those of the same protein produced endogenously in cyanobacteria. About a third of the apo-CpcA was converted to holo-CpcA. No significant bilin addition took place in a similarly engineered *E. coli* strain that lacks cpcE and cpcF. This approach should permit incisive analysis of many remaining questions in phycobiliprotein biosynthesis. These studies also demonstrate the feasibility of generating constructs of these proteins *in situ* for use as fluorescent protein probes in living cells.

ACCESSION NUMBER: 2001:482056 BIOSIS
DOCUMENT NUMBER: PREV200100482056
TITLE: Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-alpha subunit in a heterologous host.
AUTHOR(S): Tooley, Aaron J.; Cai, Yuping A.; Glazer, Alexander N. (1)
CORPORATE SOURCE: (1) Natural Reserve System, University of California System, 1111 Franklin Street, 6th Floor, Oakland, CA, 94607-5200: alexander.glazer@ucop.edu USA
SOURCE: Proceedings of the National Academy of Sciences of the United States of America, (September 11, 2001) Vol. 98, No. 19, pp. 10560-10565. print.
ISSN: 0027-8424.
DOCUMENT TYPE: Article
LANGUAGE: English

SUMMARY LANGUAGE: English

L2 ANSWER 4 OF 5 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
TI PHYCOCYANIN ALPHA-SUBUNIT PHYCOCYANOBILIN LYASE.
AB Phycobiliproteins, unlike other light-harvesting proteins involved in photosynthesis, bear covalently attached chromophores. The bilin chromophores are attached through thioether bonds to cysteine residues. The cyanobacterium *Synechococcus* sp. PCC 7002 has eight distinct bilin attachment sites on seven polypeptides, all of which carry the same chromophore, phycocyanobilin. When two genes in the phycocyanin operon of this organism, *cpcE* and *cpcF*, are inactivated by insertion, together or separately, the surprising result is elimination of correct bilin attachment at only one site, that on the .alpha. subunit of phycocyanin. We have overproduced *CpcE* and *CpcF* in *Escherichia coli*. In vitro, these proteins catalyze the attachment of phycocyanobilin to the .alpha. subunit of apophycocyanin at the appropriate site, .alpha. Cys-84, to form the correct adduct. *CpcE* and *CpcF* also efficiently catalyze the reverse reaction, in which the bilin from holo-.alpha. subunit is transferred either to the apo-.alpha. subunit of the same C-phycocyanin or to the apo-.alpha. subunit of a heterologous C-phycocyanin. The forward and reverse reactions each require both *CpcE* and *CpcF* and are specific for the .alpha.-Cys-84 position. Phycocyanobilin is the immediate precursor of the protein-bound bilin.

ACCESSION NUMBER: 1992:506225 BIOSIS
DOCUMENT NUMBER: BA94:124750
TITLE: PHYCOCYANIN ALPHA-SUBUNIT PHYCOCYANOBILIN LYASE.
AUTHOR(S): FAIRCHILD C D; ZHAO J; ZHOU J; COLSON S E; BRYANT D A;
GLAZER A N
CORPORATE SOURCE: MCB: STANLEY/DONNER ASU, 229 STANLEY HALL, UNIV. CALIF.,
BERKELEY, CALIF. 94720.
SOURCE: PROC NATL ACAD SCI U S A, (1992) 89 (15), 7017-7021.
CODEN: PNASA6. ISSN: 0027-8424.
FILE SEGMENT: BA; OLD
LANGUAGE: English

L2 ANSWER 5 OF 5 USPATFULL
TI Engineering of living cells for the expression of holo-phycobiliprotein-based constructs
AB Recombinant cells which express a fluorescent holo-phycobiliprotein fusion protein and methods of use are described. The cells comprises a bilin, a recombinant bilin reductase, an apo-phycobiliprotein fusion protein precursor of the fusion protein comprising a corresponding apo-phycobiliprotein domain, and a recombinant phycobiliprotein domain-bilin lyase, which components react to form the holo-phycobiliprotein fusion protein. Also described are holo-phycobiliprotein based transcription reporter cells and assays, which cells conditionally express a heterologous-to-the-cell, fluorescent, first holo-phycobiliprotein domain.

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

ACCESSION NUMBER: 2003:37640 USPATFULL
TITLE: Engineering of living cells for the expression of holo-phycobiliprotein-based constructs
INVENTOR(S): Glazer, Alexander N., Berkeley, CA, UNITED STATES
Tooley, Aaron J., Berkeley, CA, UNITED STATES
Cai, Yuping, Carmel, IN, UNITED STATES

	NUMBER	KIND	DATE
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PATENT INFORMATION:	US 2003027285	A1	20030206
APPLICATION INFO.:	US 2001-919486	A1	20010731 (9)
DOCUMENT TYPE:	Utility		
FILE SEGMENT:	APPLICATION		

LEGAL REPRESENTATIVE: RICHARD ARON OSMAN, SCIENCE AND TECHNOLOGY LAW GROUP,
75 DENISE DRIVE, HILLSBOROUGH, CA, 94010
NUMBER OF CLAIMS: 24
EXEMPLARY CLAIM: 1
NUMBER OF DRAWINGS: 3 Drawing Page(s)
LINE COUNT: 918
CAS INDEXING IS AVAILABLE FOR THIS PATENT.

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FILE 'MEDLINE, BIOSIS, JICST-EPLUS, FSTA, WPIDS, DGENE, USPATFULL'
ENTERED AT 16:20:18 ON 26 MAR 2003

L1 544 S C-PHYCOCYANIN
L2 5 S L1 AND HOLO-ALPHA SUBUNIT

=> s 11 and heterologous host
L3 2 L1 AND HETEROLOGOUS HOST

=> s 13 and 12
L4 2 L3 AND L2

=> d 14 ti abs ibib tot

L4 ANSWER 1 OF 2 MEDLINE
TI Biosynthesis of a fluorescent cyanobacterial C-
phycocyanin holo-alpha subunit in a
heterologous host.
AB The entire pathway for the synthesis of a fluorescent holophycobiliprotein
subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803)
was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding
enzymes required for the conversion of heme to the natural chromophore
3Z-phycyanobilin, namely, heme oxygenase 1 and 3Z-
phycyanobilin:ferredoxin oxidoreductase, were expressed from a plasmid
under control of the hybrid trp-lac (trc) promoter. Genes for the
apoprotein (C-phycocyanin alpha subunit; cpcA) and the
heterodimeric lyase (cpcE and cpcF) that catalyzes chromophore attachment
were expressed from the trc promoter on a second plasmid. Upon induction,
recombinant *E. coli* used the cellular pool of heme to produce holo-CpcA
with spectroscopic properties qualitatively and quantitatively similar to
those of the same protein produced endogenously in cyanobacteria. About a
third of the apo-CpcA was converted to holo-CpcA. No significant bilin
addition took place in a similarly engineered *E. coli* strain that lacks
cpcE and cpcF. This approach should permit incisive analysis of many
remaining questions in phycobiliprotein biosynthesis. These studies also
demonstrate the feasibility of generating constructs of these proteins in
situ for use as fluorescent protein probes in living cells.

ACCESSION NUMBER: 2001504133 MEDLINE
DOCUMENT NUMBER: 21438034 PubMed ID: 11553806

TITLE: Biosynthesis of a fluorescent cyanobacterial C-
phycocyanin holo-alpha
subunit in a heterologous host.

AUTHOR: Tooley A J; Cai Y A; Glazer A N

CORPORATE SOURCE: Department of Molecular and Cell Biology, University of
California, 142 LSA no. 3200, Berkeley, CA 94720-3200, USA.

SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE
UNITED STATES OF AMERICA, (2001 Sep 11) 98 (19) 10560-5.
Journal code: 7505876. ISSN: 0027-8424.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200111
ENTRY DATE: Entered STN: 20010913
Last Updated on STN: 20011105
Entered Medline: 20011101

L4 ANSWER 2 OF 2 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
TI Biosynthesis of a fluorescent cyanobacterial C-
phycocyanin holo-alpha subunit in a heterologous host.
AB The entire pathway for the synthesis of a fluorescent holophycobiliprotein subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803) was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to the natural chromophore 3Z-phycocyanobilin, namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase, were expressed from a plasmid under control of the hybrid trp-lac (trc) promoter. Genes for the apoprotein (C-phycocyanin alpha subunit; cpcA) and the heterodimeric lyase (cpcE and cpcF) that catalyzes chromophore attachment were expressed from the trc promoter on a second plasmid. Upon induction, recombinant *E. coli* used the cellular pool of heme to produce holo-CpcA with spectroscopic properties qualitatively and quantitatively similar to those of the same protein produced endogenously in cyanobacteria. About a third of the apo-CpcA was converted to holo-CpcA. No significant bilin addition took place in a similarly engineered *E. coli* strain that lacks cpcE and cpcF. This approach should permit incisive analysis of many remaining questions in phycobiliprotein biosynthesis. These studies also demonstrate the feasibility of generating constructs of these proteins *in situ* for use as fluorescent protein probes in living cells.

ACCESSION NUMBER: 2001:482056 BIOSIS
DOCUMENT NUMBER: PREV200100482056
TITLE: Biosynthesis of a fluorescent cyanobacterial C-
phycocyanin holo-alpha subunit in a heterologous host.
AUTHOR(S): Tooley, Aaron J.; Cai, Yuping A.; Glazer, Alexander N. (1)
CORPORATE SOURCE: (1) Natural Reserve System, University of California
System, 1111 Franklin Street, 6th Floor, Oakland, CA,
94607-5200: alexander.glazer@ucop.edu USA
SOURCE: Proceedings of the National Academy of Sciences of the
United States of America, (September 11, 2001) Vol. 98, No.
19, pp. 10560-10565. print.
ISSN: 0027-8424.
DOCUMENT TYPE: Article
LANGUAGE: English
SUMMARY LANGUAGE: English

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NEWS 9 Jun 03 New e-mail delivery for search results now available
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NEWS 11 Jun 10 PCTFULL has been reloaded
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NEWS 13 Jul 22 USAN to be reloaded July 28, 2002;
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now available on STN
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NEWS 21 Aug 19 The MEDLINE file segment of TOXCENTER has been reloaded
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NEWS 23 Sep 03 JAPIO has been reloaded and enhanced
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NEWS 25 Sep 16 CA Section Thesaurus available in CAPLUS and CA
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NEWS 30 Oct 25 MEDLINE SDI run of October 8, 2002
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NEWS 33 Dec 02 TIBKAT will be removed from STN

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NEWS 35 Dec 17 PCTFULL now covers WP/PCT Applications from 1978 to date
NEWS 36 Dec 17 TOXCENTER enhanced with additional content
NEWS 37 Dec 17 Adis Clinical Trials Insight now available on STN
NEWS 38 Dec 30 ISMEC no longer available
NEWS 39 Jan 21 NUTRACEUT offering one free connect hour in February 2003
NEWS 40 Jan 21 PHARMAML offering one free connect hour in February 2003
NEWS 41 Jan 29 Simultaneous left and right truncation added to COMPENDEX,
ENERGY, INSPEC
NEWS 42 Feb 13 CANCERLIT is no longer being updated
NEWS 43 Feb 24 METADEX enhancements
NEWS 44 Feb 24 PCTGEN now available on STN
NEWS 45 Feb 24 TEMA now available on STN

NEWS 46 Feb 26 NTIS now allows simultaneous left and right truncation
 NEWS 47 Feb 26 PCTFULL now contains images
 NEWS 48 Mar 04 SDI PACKAGE for monthly delivery of multifile SDI results
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 NEWS 50 Mar 20 EVENTLINE will be removed from STN
 NEWS 51 Mar 24 PATDPAFULL now available on STN
 NEWS 52 Mar 24 Additional information for trade-named substances without
 structures available in REGISTRY
 NEWS 53 Mar 24 Indexing from 1957 to 1966 added to records in CA/CAPLUS

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=> s phycobiliproteins
L1 1893 PHYCOBILIPROTEINS

=> s holophycobiliprotein
L2 5 HOLOPHOCOBILIPROTEIN

=> d 12 ti abs ibib tot

L2 ANSWER 1 OF 5 MEDLINE

TI Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-alpha subunit in a heterologous host.

AB The entire pathway for the synthesis of a fluorescent **holophycobiliprotein** subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803) was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to the natural chromophore 3Z-phycocyanobilin, namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase, were expressed from a plasmid under control of the hybrid trp-lac (trc) promoter. Genes for the apoprotein (C-phycocyanin alpha subunit; cpcA) and the heterodimeric lyase (cpcE and cpcF) that catalyzes chromophore attachment were expressed from the trc promoter on a second plasmid. Upon induction, recombinant *E. coli* used the cellular pool of heme to produce holo-CpcA with spectroscopic properties qualitatively and quantitatively similar to those of the same protein produced endogenously in cyanobacteria. About a third of the apo-CpcA was converted to holo-CpcA. No significant bilin addition took place in a similarly engineered *E. coli* strain that lacks cpcE and cpcF. This approach should permit incisive analysis of many remaining questions in phycobiliprotein biosynthesis. These studies also demonstrate the feasibility of generating constructs of these proteins *in situ* for use as fluorescent protein probes in living cells.

ACCESSION NUMBER: 2001504133 MEDLINE

DOCUMENT NUMBER: 21438034 PubMed ID: 11553806

TITLE: Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-alpha subunit in a heterologous host.

AUTHOR: Tooley A J; Cai Y A; Glazer A N

CORPORATE SOURCE: Department of Molecular and Cell Biology, University of California, 142 LSA no. 3200, Berkeley, CA 94720-3200, USA.

SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (2001 Sep 11) 98 (19) 10560-5.
Journal code: 7505876. ISSN: 0027-8424.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200111

ENTRY DATE: Entered STN: 20010913

Last Updated on STN: 20011105

Entered Medline: 20011101

L2 ANSWER 2 OF 5 USPATFULL

TI Engineering of living cells for the expression of holo-phycobiliprotein-based constructs

AB Recombinant cells which express a fluorescent holo-phycobiliprotein

fusion protein and methods of use are described. The cells comprises a bilin, a recombinant bilin reductase, an apo-phycobiliprotein fusion protein precursor of the fusion protein comprising a corresponding apo-phycobiliprotein domain, and a recombinant phycobiliprotein domain-bilin lyase, which components react to form the holo-phycobiliprotein fusion protein. Also described are holo-phycobiliprotein based transcription reporter cells and assays, which cells conditionally express a heterologous-to-the-cell, fluorescent, first holo-phycobiliprotein domain.

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

ACCESSION NUMBER: 2003:37640 USPATFULL
TITLE: Engineering of living cells for the expression of holo-phycobiliprotein-based constructs
INVENTOR(S): Glazer, Alexander N., Berkeley, CA, UNITED STATES
Tooley, Aaron J., Berkeley, CA, UNITED STATES
Cai, Yuping, Carmel, IN, UNITED STATES

	NUMBER	KIND	DATE
PATENT INFORMATION:	US 2003027285	A1	20030206
APPLICATION INFO.:	US 2001-919486	A1	20010731 (9)
DOCUMENT TYPE:	Utility		
FILE SEGMENT:	APPLICATION		
LEGAL REPRESENTATIVE:	RICHARD ARON OSMAN, SCIENCE AND TECHNOLOGY LAW GROUP, 75 DENISE DRIVE, HILLSBOROUGH, CA, 94010		
NUMBER OF CLAIMS:	24		
EXEMPLARY CLAIM:	1		
NUMBER OF DRAWINGS:	3 Drawing Page(s)		
LINE COUNT:	918		

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

L2 ANSWER 3 OF 5 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.
TI Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-.alpha. subunit in a heterologous host.
AB The entire pathway for the synthesis of a fluorescent holophycobiliprotein subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803) was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to the natural chromophore 3Z-phycocyanobilin, namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase, were expressed from a plasmid under control of the hybrid trp-lac (trc) promoter. Genes for the apoprotein (C-phycocyanin .alpha. subunit; cpcA) and the heterodimeric lyase (cpcE and cpcF) that catalyzes chromophore attachment were expressed from the trc promoter on a second plasmid. Upon induction, recombinant *E. coli* used the cellular pool of heme to produce holo-CpcA with spectroscopic properties qualitatively and quantitatively similar to those of the same protein produced endogenously in cyanobacteria. About a third of the apo-CpcA was converted to holo-CpcA. No significant bilin addition took place in a similarly engineered *E. coli* strain that lacks cpcE and cpcF. This approach should permit incisive analysis of many remaining questions in phycobiliprotein biosynthesis. These studies also demonstrate the feasibility of generating constructs of these proteins *in situ* for use as fluorescent protein probes in living cells.

ACCESSION NUMBER: 2001329835 EMBASE
TITLE: Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-.alpha. subunit in a heterologous host.
AUTHOR: Tooley A.J.; Cai Y.A.; Glazer A.N.
CORPORATE SOURCE: A.N. Glazer, Natural Reserve System, University of California System, 1111 Franklin Street, Oakland, CA 94607-5200, United States. alexander.glazer@ucop.edu
SOURCE: Proceedings of the National Academy of Sciences of the United States of America, (11 Sep 2001) 98/19 (10560-10565).

Refs: 30
ISSN: 0027-8424 CODEN: PNASA6
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English

L2 ANSWER 4 OF 5 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
TI Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-alpha subunit in a heterologous host.
AB The entire pathway for the synthesis of a fluorescent **holophycobiliprotein** subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803) was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to the natural chromophore 3Z-phycocyanobilin, namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase, were expressed from a plasmid under control of the hybrid trp-lac (trc) promoter. Genes for the apoprotein (C-phycocyanin alpha subunit; cpcA) and the heterodimeric lyase (cpcE and cpcF) that catalyzes chromophore attachment were expressed from the trc promoter on a second plasmid. Upon induction, recombinant *E. coli* used the cellular pool of heme to produce holo-CpcA with spectroscopic properties qualitatively and quantitatively similar to those of the same protein produced endogenously in cyanobacteria. About a third of the apo-CpcA was converted to holo-CpcA. No significant bilin addition took place in a similarly engineered *E. coli* strain that lacks cpcE and cpcF. This approach should permit incisive analysis of many remaining questions in phycobiliprotein biosynthesis. These studies also demonstrate the feasibility of generating constructs of these proteins *in situ* for use as fluorescent protein probes in living cells.

ACCESSION NUMBER: 2001:482056 BIOSIS
DOCUMENT NUMBER: PREV200100482056
TITLE: Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-alpha subunit in a heterologous host.
AUTHOR(S): Tooley, Aaron J.; Cai, Yuping A.; Glazer, Alexander N. (1)
CORPORATE SOURCE: (1) Natural Reserve System, University of California System, 1111 Franklin Street, 6th Floor, Oakland, CA, 94607-5200: alexander.glazer@ucop.edu USA
SOURCE: Proceedings of the National Academy of Sciences of the United States of America, (September 11, 2001) Vol. 98, No. 19, pp. 10560-10565. print.
ISSN: 0027-8424.
DOCUMENT TYPE: Article
LANGUAGE: English
SUMMARY LANGUAGE: English

L2 ANSWER 5 OF 5 HCAPLUS COPYRIGHT 2003 ACS
TI Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-.alpha. subunit in a heterologous host
AB The entire pathway for the synthesis of a fluorescent **holophycobiliprotein** subunit from a photosynthetic cyanobacterium (*Synechocystis* sp. PCC6803) was reconstituted in *Escherichia coli*. Cyanobacterial genes encoding enzymes required for the conversion of heme to the natural chromophore 3Z-phycocyanobilin, namely, heme oxygenase 1 and 3Z-phycocyanobilin:ferredoxin oxidoreductase, were expressed from a plasmid under control of the hybrid trp-lac (trc) promoter. Genes for the apoprotein (C-phycocyanin .alpha. subunit; cpcA) and the heterodimeric lyase (cpcE and cpcF) that catalyzes chromophore attachment were expressed from the trc promoter on a second plasmid. Upon induction, recombinant *E. coli* used the cellular pool of heme to produce holo-CpcA with spectroscopic properties qual. and quant. similar to those of the same protein produced endogenously in cyanobacteria. About a third of the apo-CpcA was converted to holo-CpcA. No significant bilin addn. took place in a similarly engineered *E. coli* strain that lacks cpcE and cpcF.

This approach should permit incisive anal. of many remaining questions in phycobiliprotein biosynthesis. These studies also demonstrate the feasibility of generating constructs of these proteins *in situ* for use as fluorescent protein probes in living cells.

ACCESSION NUMBER: 2001:705481 HCPLUS
DOCUMENT NUMBER: 136:2664
TITLE: Biosynthesis of a fluorescent cyanobacterial C-phycocyanin holo-.alpha. subunit in a heterologous host
AUTHOR(S): Tooley, Aaron J.; Cai, Yuping A.; Glazer, Alexander N.
CORPORATE SOURCE: Department of Molecular and Cell Biology, University of California, Berkeley, CA, 94720-3200, USA
SOURCE: Proceedings of the National Academy of Sciences of the United States of America (2001), 98(19), 10560-10565
CODEN: PNASA6; ISSN: 0027-8424
PUBLISHER: National Academy of Sciences
DOCUMENT TYPE: Journal
LANGUAGE: English
REFERENCE COUNT: 30 THERE ARE 30 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

=> d his

(FILE 'HOME' ENTERED AT 15:45:53 ON 26 MAR 2003)

FILE 'MEDLINE, USPATFULL, DGENE, EMBASE, JICST-EPLUS, FSTA, WPIDS, BIOSIS, BIOBUSINESS, JAPIO, HCPLUS' ENTERED AT 15:48:26 ON 26 MAR 2003

L1 1893 S PHYCOBILIPROTEINS
L2 5 S HOLOPHYCOBILIPROTEIN

=> s apohycobiliprotein
L3 0 APOHYCOBILIPROTEIN

=> s fusion protein
L4 139090 FUSION PROTEIN

=> s heterologous
L5 184332 HETEROLOGOUS

=> s 15 and 14
L6 18399 L5 AND L4

=> s 16 and 12
L7 1 L6 AND L2

=> d 17 ti abs ibib tot

L7 ANSWER 1 OF 1 USPATFULL
TI Engineering of living cells for the expression of holo-phycobiliprotein-based constructs

AB Recombinant cells which express a fluorescent holo-phycobiliprotein fusion protein and methods of use are described. The cells comprises a bilin, a recombinant bilin reductase, an apo-phycobiliprotein fusion protein precursor of the fusion protein comprising a corresponding apo-phycobiliprotein domain, and a recombinant phycobiliprotein domain-bilin lyase, which components react to form the holo-phycobiliprotein fusion protein. Also described are holo-phycobiliprotein based transcription reporter cells and assays, which cells conditionally express a heterologous-to-the-cell, fluorescent, first holo-phycobiliprotein domain.

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

ACCESSION NUMBER: 2003:37640 USPATFULL
TITLE: Engineering of living cells for the expression of holo-phycobiliprotein-based constructs
INVENTOR(S): Glazer, Alexander N., Berkeley, CA, UNITED STATES
Tooley, Aaron J., Berkeley, CA, UNITED STATES
Cai, Yuping, Carmel, IN, UNITED STATES

	NUMBER	KIND	DATE
PATENT INFORMATION:	US 2003027285	A1	20030206
APPLICATION INFO.:	US 2001-919486	A1	20010731 (9)
DOCUMENT TYPE:	Utility		
FILE SEGMENT:	APPLICATION		
LEGAL REPRESENTATIVE:	RICHARD ARON OSMAN, SCIENCE AND TECHNOLOGY LAW GROUP, 75 DENISE DRIVE, HILLSBOROUGH, CA, 94010		
NUMBER OF CLAIMS:	24		
EXEMPLARY CLAIM:	1		
NUMBER OF DRAWINGS:	3 Drawing Page(s)		
LINE COUNT:	918		

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

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(FILE 'HOME' ENTERED AT 15:45:53 ON 26 MAR 2003)

FILE 'MEDLINE, USPATFULL, DGENE, EMBASE, JICST-EPLUS, FSTA, WPIDS,
BIOSIS, BIOPARTNERS, JAPIO, HCAPLUS' ENTERED AT 15:48:26 ON 26 MAR 2003

L1 1893 S PHYCOBILIPROTEINS
L2 5 S HOLOPHYCOBILIPROTEIN
L3 0 S APOHYCOBILIPROTEIN
L4 139090 S FUSION PROTEIN
L5 184332 S HETEROLOGOUS
L6 18399 S L5 AND L4
L7 1 S L6 AND L2

=> s recombinant cell
L8 10678 RECOMBINANT CELL

=> s 18 and protein expression
8 FILES SEARCHED...
L9 2727 L8 AND PROTEIN EXPRESSION

=> s 19 and 12
L10 0 L9 AND L2

=> s 19 and 11
L11 1 L9 AND L1

=> s 111 not 17
L12 1 L11 NOT L7

=> d 111 ti abs ibib tot

L11 ANSWER 1 OF 1 USPATFULL
TI Recombinant phycobiliprotein and phycobiliprotein linker fusion proteins and uses therefore
AB This invention is directed to the utilization of the developing methods for molecular manipulation of cyanobacteria and red algae (and potentially cryptomonad algae) to express of phycobiliproteins and phycobiliprotein linker fusion proteins and their utilization as phycobiliprotein, phycobilisome and subassembly based reagents. In particular, the present invention relates to a method for a specific binding assay to determine a target moiety which is a member of a

specific binding pair, and provides an improvement in the method comprising using a detectable label which is a fusion protein containing both a phycobiliprotein domain and another domain corresponding to a first member of a specific binding pair, where the fusion protein binds to a second member of the specific binding pair to provide a detectable labeled complex. The domain derived from the first member of the specific binding pair can be directly fused to the phycobiliprotein or phycobiliprotein linker domain or be separated by a spacer that allows correct folding of both domains.

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

ACCESSION NUMBER: 2001:237667 USPATFULL
TITLE: Recombinant phycobiliprotein and phycobiliprotein linker fusion proteins and uses therefore
INVENTOR(S): Allnutt, F.C. Thomas, Port Deposit, MD, United States
Toole, Colleen Mary, New Winson, MD, United States
Morseman, John Peter, Columbia, MD, United States

	NUMBER	KIND	DATE
PATENT INFORMATION:	US 2001055783	A1	20011227
APPLICATION INFO.:	US 2001-882093	A1	20010618 (9)

	NUMBER	DATE
PRIORITY INFORMATION:	US 2000-211784P	20000616 (60)
DOCUMENT TYPE:	Utility	
FILE SEGMENT:	APPLICATION	
LEGAL REPRESENTATIVE:	BROBECK, PHLEGER & HARRISON, LLP, ATTN: INTELLECTUAL PROPERTY DEPARTMENT, 1333 H STREET, N.W. SUITE 800, WASHINGTON, DC, 20005	
NUMBER OF CLAIMS:	46	
EXEMPLARY CLAIM:	1	
LINE COUNT:	1218	

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

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FILE 'MEDLINE, USPATFULL, DGENE, EMBASE, JICST-EPLUS, FSTA, WPIDS, BIOSIS, BIOPARTNERS, JAPIO, HCAPLUS' ENTERED AT 15:48:26 ON 26 MAR 2003

L1 1893 S PHYCOBILIPROTEINS
L2 5 S HOLOPHYCOBILIPROTEIN
L3 0 S APOHYCOBILIPROTEIN
L4 139090 S FUSION PROTEIN
L5 184332 S HETEROLOGOUS
L6 18399 S L5 AND L4
L7 1 S L6 AND L2
L8 10678 S RECOMBINANT CELL
L9 2727 S L8 AND PROTEIN EXPRESSION
L10 0 S L9 AND L2
L11 1 S L9 AND L11
L12 1 S L11 NOT L7

=> s bilioprotein
L13 734 BILIOPROTEIN

=> s l13 and l8
L14 1 L13 AND L8

=> s l9 and l13
L15 1 L9 AND L13

=> d 115 ti abs ibib tot

L15 ANSWER 1 OF 1 USPATFULL

TI Recombinant phycobiliprotein and phycobiliprotein linker fusion proteins and uses therefore

AB This invention is directed to the utilization of the developing methods for molecular manipulation of cyanobacteria and red algae (and potentially cryptomonad algae) to express of phycobiliproteins and phycobiliprotein linker fusion proteins and their utilization as phycobiliprotein, phycobilisome and subassembly based reagents. In particular, the present invention relates to a method for a specific binding assay to determine a target moiety which is a member of a specific binding pair, and provides an improvement in the method comprising using a detectable label which is a fusion protein containing both a phycobiliprotein domain and another domain corresponding to a first member of a specific binding pair, where the fusion protein binds to a second member of the specific binding pair to provide a detectable labeled complex. The domain derived from the first member of the specific binding pair can be directly fused to the phycobiliprotein or phycobiliprotein linker domain or be separated by a spacer that allows correct folding of both domains.

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

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TITLE: Recombinant phycobiliprotein and phycobiliprotein linker fusion proteins and uses therefore

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